

UNIVERSITY OF ARKANSAS
COLLEGE OF AGRICULTURE

Agricultural Experiment Station

Main Station, University; with Cotton Branch Station, Lee County
Rice Branch Station, Arkansas County; Fruit and
Truck Branch Station, Hempstead County

Further Studies on the Overwintering and Dissemination of the Fire-Blight Pathogen

H. R. ROSEN

BULLETIN NO. 283

DAN T. GRAY, Director

FAYETTEVILLE, ARKANSAS

MAY, 1933

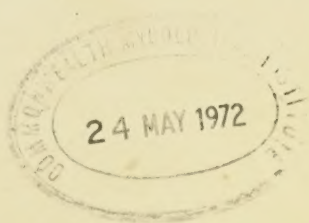


TABLE OF CONTENTS

	Page
Studies of overwintered cankers of apples and pears in relation to blight development	4
Current pruning practices in relation to blight development	11
Studies on the succession of fire blight	14
The relationship of weather conditions to the development of blight	19
Sequence of fire blight within a growing season	29
First blight on pears	29
First blight on apples	43
Primary twig blight on apples and pears	47
Insects as carriers and disseminators of the fire-blight pathogen	51
Investigations on honeybees	51
Investigations on other insects as blight distributors ...	67
Twig infections as related to the presence or absence of insects	71
Attempts to control fire blight by means of sprays	82
Summary and conclusions	96
List of references	99

FURTHER STUDIES ON THE OVERWINTERING AND DISSEMINATION OF THE FIRE-BLIGHT PATHOGEN

By H. R. ROSEN

Department of Plant Pathology

The manner in which the fire-blight pathogen lives from one year to another and the agents concerned in its dissemination constitute for the present the most outstanding problems in the prevention of the fire-blight disease of apples, pears, quinces, and other plants. Without a proper knowledge of the underlying natural phenomena involved in these processes there can be no certainty in any practice which aims to control this destructive malady.

In view of the fact that pear trees had been considered as the chief host in which the fire-blight bacteria are able to overwinter in the Ozarks of Arkansas (1), the first investigations were largely concerned in determining the adequacy of this theory. If it were true, then the removal or destruction of pears should at once result in a very material reduction of the disease on apples, or in its complete suppression. Unfortunately, however, the initial investigations did not substantiate this theory (29).

It was found that fire-blight bacteria could be isolated from blighted twigs and limbs of apples throughout the winter season just as readily as from pears; that such isolations, in the form of pure cultures, resulted in typical fire-blight infections when they were artificially introduced into healthy pear and apple shoots; and that field studies during the growing seasons of 1927 to 1929 suggested in general no relationship between the prevalence and severity of blight on apples and that on pears. However, it was found that since the blossoming and growing period of the common varieties of pears grown in the Ozarks precedes that of apples, blight is likely to appear first on pears and then spread to nearby apples if conditions are proper.

Despite the fact that no great difficulty was encountered in finding the fire-blight parasite, *Erwinia amylovora*, in blighted wood of pears and apples during the winter and spring seasons, the detection of the germ in the early spring on such material in the form of droplets of exudate was reported to be a complete failure. The commonly accepted theory that the bacterial exudations from overwintered blight cankers constitute the source of inoculum for the renewal of spring blight, was not substantiated.

Although the studies previously reported (29) on the behavior of overwintered cankers involved hundreds of marked cankers on both apples and pears, and had extended over a 3-year period, nevertheless, it appeared desirable to continue

this line of investigation in order to make sure that exudations as the source of the first spring blight had not been overlooked. The studies that are now to be reported include, therefore, a 3-year additional period of field observations and laboratory studies of overwintered cankers, and their exudates on both apple and pear. They likewise include a somewhat extensive study of the nature of the first spring blight, its relationship to overwintered cankers, the weather conditions associated with it, the possible role of insects as disseminating agents of the causal organism, and attempts to control it by utilizing the facts deduced from the studies. A study of secondary, or later blight, has also been made with reference to origin, weather conditions and insects associated with it, and its possible control.

STUDIES OF OVERWINTERED CANCERS OF APPLES AND PEARS IN RELATION TO BLIGHT DEVELOPMENT

Since the investigations reported in the previous publication (29) dealt mainly with the material and observations of only 5 acres of apples and about 10 acres of pears, it seemed desirable to enlarge the area and extent of observations. With this end in view, an attempt was made to survey all apple and pear orchards in Washington and Benton counties, the two principal apple growing counties in Arkansas, which contained susceptible varieties. These field studies were conducted during the years 1929 to 1932,¹ which—in view of the 1930 fire-blight epidemic, one of the worst that has been experienced by Arkansas and Missouri apple growers—made possible a very large amount of material for study. In these years the total number of blighted twigs, blossom clusters, and overwintered cankers that have been included in these observations is not known, but over 3,000 acres were scrutinized, involving about 90,000 apple trees and most of the pear trees (several hundred)² located in the main apple growing townships of these two counties. In so far as amount of diseased material, length of observations, and extent of areas involved, the writer knows of no similar study on this disease.

The necessity for studying the disease in a somewhat wide area, while possessing certain marked disadvantages nevertheless seemed quite essential. There is no certainty that, given any one or two orchards, blight will be present in successive years in sufficient amount to enable an adequate study of the disease.

Considering the influence of diverse climatic and soil con-

¹The writer acknowledges the able assistance of Glenn Boyd, Aubrey Gates, and Ross Mauney in these field studies.

²This region at one time contained a relatively large number of commercial pear orchards, but owing to the ravages of fire blight and the ruling in 1922 of the State Plant Board, prohibiting the growing of pears in predominant apple growing townships, very few pear orchards are now left.

ditions on plant diseases as a whole, attention is called to the chief characteristics of the two apple-growing counties included in these studies. Both counties are located in the Ozark Mountain region, Benton County representing mainly a plateau region of about 1300 feet in elevation, with Washington County much more rolling or hilly in parts, though as a whole of comparable elevation. The soil is mainly derived from limestone and shale, with silt loams predominating in both counties. The combined land area of the two is about 1,171,840 acres³ and the farm areas including apple orchards consist of 870,923 acres,³ the remainder being largely made up of hardwood forests. The predominant apple growing townships, while more or less contiguous in Benton County, are somewhat widely scattered in Washington County, and in many instances commercial orchards containing varieties of apples susceptible to fire blight are considerable distances removed from other similar orchards.

The climate of the region, compared with northern fruit-growing areas, is characterized by a long growing season, beginning in March and ending in November as far as pomaceous plants are concerned. The alternate freezing and thawing of night and day temperatures is common during the winter and early spring, and is almost of daily occurrence during the late winter and early spring. Even in midwinter, temperatures below freezing rarely continue for more than a few days and are followed by warm sunny days such as northern fruit regions rarely experience. Both in summer and winter this region is characterized by marked diurnal fluctuations in temperature, hot days and cool nights in the summer, and bright pleasant days succeeded by frosty nights, interspersed between more or less severe cold periods, in the winter. The annual rainfall of about 42 inches is fairly well distributed throughout the year, being somewhat less during the winter months than in the growing season. Snowfall as a whole is rare, and snow seldom lasts more than a few days.

Sap flow from wounded or diseased portions of trees is likely to occur throughout most of the winter season, clearly suggesting a more or less continued root activity and the development of hydrostatic pressure. This recurrent physiological activity during the winter season, so unlike that of northern regions where the soil remains frozen for considerable depths, is likely to influence bacterial exudations. Such exudations, according to the common theory, presumably arise from pressure exerted by bacterial growths within the tissues, forcing out the bacteria through natural openings or through rifts in the tissues. Why such pressure should result in outward expulsion and not in inward expansion of strands within the air spaces, has not

³Fifteenth United States Census: 1930. Agriculture Vol. 2, Part 2, 1609 pp. 1932.

been explained. That exudations cannot always be considered as due merely to the continued growth of bacteria, may readily be proved by placing fire-blight-infected pear, apple, or rose shoots, which show no exudates, into moist chambers and subjecting them to low temperatures of a refrigerator. When this is done, bacterial droplets of ooze are likely to appear in great numbers on the surface of diseased areas, suggesting that under such conditions changes in temperature and moisture resulting in changes of hydrostatic and pneumatic pressures exerted within host tissues probably have some influence on the expulsion of the bacteria. To the writer, a more reasonable explanation for the occurrence of bacterial exudations may be sought in the abnormal internal pressure, more or less localized and corresponding to the region of maximum bacterial invasion, brought about by the stoppage, or interference, of gaseous exchange within the air chambers of cortical tissues by the plugging of the pneumatic system with the thickly-populated bacterial strands.

According to this theory one may expect bacterial exudations to be more or less concomittant with the occurrence of sap flow, and consequently, in those sections of the country where such flow is completely stopped during the winter season, the bacterial exudations may be expected to appear in great quantities as soon as the soil thaws out sufficiently to enable the roots to function and to generate hydrostatic pressure. On the other hand, in Arkansas, and probably in other states where the roots of trees are more or less active throughout the winter, bacterial exudations from active cankers theoretically should appear whenever sap flow occurs, and should not be limited to the spring and early summer seasons. At any rate, it is obvious that the marked differences in climatic conditions between northern and southern fruit-growing sections, resulting in marked differences in physiological responses of the trees, do not warrant the assumption that overwintered blight and bacterial exudations would respond in the same way in the different sections.

For studying the behavior of overwintered cankers and blighted twigs, three different criteria were used: first, signs of extension of canker margins during the winter and spring; second, the production of bacterial ooze or exudate; and third, attempts to isolate the parasite from the interiors of blighted wood, particularly from the portions joining the healthy wood. The results of these studies are as follows:

On Kieffer pears, and the apple varieties of Jonathan, Maiden Blush, Yellow Transparent, and Ada Red, signs of fresh extensions of the margins of overwintered cankers were relatively rare. Out of every 200-lot sample of blighted twigs and limbs, one twig, on the average, would show an extension of the canker in the form of a darker, less withered, diseased area. More-

over, these fresh extensions, which usually made their appearance in late February and early March on pears, and in late March and early April on apples, were in many instances unassociated with the presence of infectious fire-blight bacteria. When the tissues involved in such extensions were surface sterilized, mashed in sterile water, and injected by means of hypodermic needles into healthy pear shoots maintained in a greenhouse, only one out of four produced typical fire-blight infections. The fact that the older blighted parts were in most instances studded with fungus-fruited bodies, suggests that the newer extensions were due largely either to the renewed activity of secondary invaders, or to some internal injury, unassociated with *Erwinia amylovora*, which may have occurred prior to the appearance of the external symptoms.

Since most twig and limb infections were found to occur prior to July 15, and since late summer or fall infections were found to be rare, it is apparent that a period of about $4\frac{1}{2}$ months elapses between the cessation of most twig and limb blight and the advent of the dormant season. A period of such length, which is almost equivalent to the whole of the apple-growing season in southern Wisconsin, enables secondary invaders to become well established in blighted wood, and possibly to renew their activities in the following spring.

During the months of February, March, and April a very thorough search was made for fresh bacterial exudates from overwintered material. A conservative estimate of the number of blighted twigs and limbs carefully examined each year for the 3-year period would be between 15,000 and 20,000. Of this number, 80 per cent or more were represented by blighted twigs less than one-half inch in diameter, 15 per cent by limbs up to $1\frac{1}{2}$ inches in diameter, and the remainder mostly on pear, but occasionally on apple, by limbs and trunks of larger diameter. Many of these were examined at least once a week during the months mentioned. Several hundred blighted twigs and limbs on pears and apples located within short walking distance of the laboratory were examined daily for signs of oozing. Altogether the number of oozing limb and twig cankers found during the 3-year period does not exceed 25. In each case, the droplet of ooze (see Figure 1) was removed from the blighted organ, diluted slightly with sterile water, and injected into healthy succulent pear shoots. No infections were obtained. On the other hand, no difficulty was experienced in obtaining infections under the same conditions when either pure cultures were used or when contaminated honeycomb material was used. In some instances bacteria were observed under the microscope when a small portion of the ooze was mounted in water in a hanging-drop slide, but these bacteria were apparently not infectious. When the diluted ooze was plated out in a series of Petri dishes, most of

the bacterial colonies which appeared were yellowish and representative of saprophytes which are commonly obtained from old diseased material.

Notwithstanding the fact that no fresh, infectious exudations were located prior to the first signs of blight, there was no



Figure 1. Droplet of exudate, near the center, on Kieffer pear twig gathered on April 11, 1932, near Bentonville. This exudate, typical of that observed prior to the first signs of fresh blight, was located in a twig area which had previously been blighted, but the bacteria present in the droplet failed to produce infections.

difficulty in obtaining typical fire-blight infections and pure cultures of *Er. amylovora* from blighted twigs and limbs of apple and pear throughout the winter and spring, provided a sufficient number was used. Given 100 blighted twigs, infectious fire-blight bacteria can always be isolated from one or two twigs. On larger limbs the writer has insufficient data to indicate the percentage of overwintering, since the number investigated, some 200 in all over a period of 6 years, is too small. On apples, fire blight in Arkansas rarely involves limbs that are more than 1½ inches in diameter on Jonathan, Maiden Blush, Yellow Transparent, Grimes Golden, and Ada Red, the most common susceptible varieties grown in this state. The data gathered on this phase of investigation, were comparable to those previously published (29, pp. 12-31).

Briefly summarizing the investigations on the behavior of overwintered cankers and blighted twigs, it appears that the data gathered in the years 1930, 1931, and 1932 are very similar to those obtained in the years 1927, 1928 and 1929. In this 6-year period, there is reasonable certainty that fresh, infectious exudations from overwintered material were not concerned with the production of first spring blight in Arkansas. How does this compare with the findings in other sections of America?

Having previously presented a summary of the literature to 1929 upon the behavior of overwintered material (29), it is desirable to discuss the more recent studies on this subject in order to determine the points of similarity and disagreement, and possible explanations for any discrepancies. The most important studies are those by Miller (20) and by Pierstorff (23). Miller, working in southern Wisconsin, found that 6 per cent of the cankers examined on Fameuse apple trees in 1926 overwintered the bacteria, 7 per cent acted as hold-over blight in 1927, and 3 per cent in 1928. Other varieties showed a smaller percentage, Wealthy with 2 per cent in 1926, 2 per cent in 1927, 1 per cent in 1928; and Dudley and McIntosh with very few or no overwintering cankers. The criteria used in determining the number of cases in which overwintering occurred, consisted in signs of the extension of margins in the spring, and the production of exudate from the surface of the discolored areas. While these would not be adequate in Arkansas in view of the fact that spring exudates from fire-blight cankers have been found here to be non-infectious, they may be adequate under Wisconsin conditions. Considering the marked differences in climatic conditions in the two sections, with long hot summers, short, relatively dry dormant seasons, short warm days, and cool to frosty nights during the spring in Arkansas; and the short summers, long wet dormant season with longer days and more even day and night temperatures during the early parts of the growing season in southern Wisconsin, one may expect considerable differ-

ence in the behavior of overwintered cankers. These climatic differences, coupled with differences in varieties grown in the two sections, the latter being a point which in the writer's judgment Miller has properly emphasized, may be expected to yield diverse reactions in both host and parasite. Some of the differences in climatic conditions prevalent in the two localities during the early part of the apple growing season may be seen in Figures 4 and 5.

Pierstorff's studies in New York, on the behavior of overwintered cankers, while not quite as extensive as Miller's, are of interest. Observations made of hold-over blight at blossoming time in an apple orchard near Ithaca in 1925 and 1926 showed that out of 158 branches of diameters varying from one-fourth to three-fourths of an inch of King apple, 27, or about 17 per cent, contained viable bacteria in 1925, while in 1926 somewhat over 10 per cent of the branches of this variety contained viable bacteria. In the McIntosh variety slightly over one per cent were found to be holdovers in 1925, and 2.6 per cent in 1926. The percentage of overwintering found by Pierstorff in Tompkins King is surprisingly high, and if it were used as a measure in determining the relative prevalence or severity of blight in New York compared with that in Arkansas, it would lead to erroneous conclusions. It is to be noted that fire blight on both apples and pears is far more common and destructive in the South as a whole, including Arkansas, than it is in the northern states, as shown in various Supplements of the Plant Disease Reporter, issued by the Office of Mycology and Disease Survey of the United States Department of Agriculture. These reports indicate that the disease is often three to five times as abundant in Arkansas as in New York. Concerning the production of spring exudate on overwintered material, Pierstorff is apparently of the opinion that such exudations are responsible for the initiation of blight, although he makes no positive statement to this effect. He says (23, p. 29), "From the foregoing data (referring to spring isolations from overwintered apple branches) it is evident that small twigs may serve as hold-overs for fire blight. In Plate II, three drops of ooze which are sometimes found on these small twigs are shown."

It thus appears that as far as the presence of *Er. amylovora* within overwintered small and large blighted branches of apple are concerned, the investigations in Wisconsin, New York, and Arkansas are in general agreement, although there is considerable divergence in the percentage of overwintering. However, concerning the production of spring exudate from overwintered cankers and its possible role as inoculum for early blight production, there is no apparent agreement. It is thus possible that either the observations in Arkansas or elsewhere are faulty, or that there is a marked difference in the behavior of hold-over

blight in the early part of the growing season in different sections of the country. There is close agreement concerning the ability of relatively small, blighted apple twigs to carry the pathogen within the tissues through the winter, and a similar agreement in postulating that it is unnecessary to seek for large limb cankers and body cankers to explain successful overwintering in all cases.

CURRENT PRUNING PRACTICES IN RELATION TO BLIGHT DEVELOPMENT

Irrespective of the presence or absence of fire blight, one of the standard practices in almost all commercial orchards of the Ozarks has for many years consisted of a more or less severe winter or early spring pruning. The theory behind this practice was that such pruning would increase the quality of the fruit, the productivity of the trees, and produce a form of growth conducive to long life and economy of production. While this theory has been questioned in recent years, pruning of apples still remains a common practice, though the amount is probably not as great as it used to be. In the course of this general pruning, dead and diseased wood, including branches affected by fire blight, are removed. When blighted limbs are numerous, the amount of pruned wood is often so great as to leave but a skeleton outline of the tree (Figure 2).

So severe is the pruning at times, made necessary by the severity of blight, that nearly all bearing wood is removed (compare Figure 2 with Figure 3) and the large remaining branches become subject to sunscald on the upper surfaces. These sunscalded areas, brought about mainly by the exposure to direct sunlight, of limbs that had been more or less shaded previously by smaller foliar shoots, in turn become the seat of invasion by saprophytes or very weak parasites, such as *Schizophyllum*. This also adds to the amount of wood rendered barren and rotted. As far as pruning out blighted branches is concerned, this has been a very common winter and early spring practice in nearly all commercial apple orchards of susceptible varieties. This of course, has by no means resulted in the complete removal of all blighted wood. No matter how careful a fruit-grower may be in removing blighted parts and taking pains to cut some distance below the diseased areas, it is practically impossible to detect and remove all blighted wood on trees that have a large number of blighted twigs and limbs. As a whole, large diseased branches, being more readily observable than small blighted twigs, are more likely to be pruned out, but when a tree has over 500 blighted twigs and limbs (29, p. 26) as occasionally happens, then even with the utmost care, a few blighted branches are likely to be overlooked. And when the pruning is not carefully



Figure 2. A Jonathan tree which was so severely blighted in 1930 that practically all the bearing wood was removed in the pruning operation. Photographed at Farmington, April 14, 1931. Compare with Figure 3.

done, as many as one-fourth of the blighted twigs may be left. Usually, however, the number of blighted branches left on the trees would not exceed one per cent of the former year's blight, and in many orchards it is less than that, in accordance with the writer's observations.

What is the relationship between pruning practices in Arkansas and overwintering of the pathogen? According to the prevailing belief, overwintering occurs entirely within the tissues of unpruned, blighted trunks, limbs, twigs, and roots, the roots being considered the main seat of overwintering in Oregon (25). As blight of the main trunks is extremely rare in Arkan-

sas, and as root blight and collar blight has not been associated with the activities of *Er. amylovora* in this state, then limbs and twigs must be the only seat of overwintering, if one accepts the current belief. Now, as the writer's studies indicate that it usually requires a lot of about 100 blighted limbs and twigs to find



Figure 3. A Jonathan tree, standing near the one shown in Figure 2. Photographed at Farmington, April 14, 1931, when the other tree had no blossoms. This tree had very little blight in 1930.

one case of successful overwintering within the tissues, it follows that in those orchards where blight was not present in the previous season, in those where blight was limited to only a few twigs or flower clusters, and in those where pruning has removed all but a few undetected blighted branches, the possibility of the pathogen being present the following season would be very slight. Therefore, fire blight in those orchards would not appear unless the pathogen were brought in from some source outside of orchards, or unless overwintering, contrary to the current theory, is accomplished in some additional ways.

Is fire blight restricted to those orchards which exhibited the disease in the previous season or to those which happen to be close to blighted trees?

STUDIES ON THE SUCCESSION OF FIRE BLIGHT

The studies on the succession of fire blight in a large number of orchards were favored by the severe epidemic of blight on apples which occurred in 1930. As the field studies on 92 apple orchards and two pear orchards in 1929 were continued in the same orchards in 1930, it happened that an exceptional opportunity presented itself for studying the succession of blight.

In a previous publication (29, *p. 16*) it was noted that in 1929, blight was present only in moderate amounts on apples as a whole and was more common and severe on pears. Of the 92 apple orchards observed in both 1929 and 1930, the record for 1929 was as follows: 19 orchards apparently had no blight, 43 showed a trace of blight, a trace being considered from one to several blighted twigs or flower clusters over a whole orchard, 16 had a slight amount of blight, varying from a fraction up to 5 per cent of the twigs or flower clusters, 9 possessed a medium amount of blight, from 6 to 10 per cent blossom or twig blight, and 5 had considerable blight, varying from 11 to 30 per cent of blossoms or twigs. Of 105 additional apple orchards under observation in 1929, but not in 1930, 29 orchards had no blight, 45 possessed a trace, 29 showed a slight amount, and two had more than 6 per cent blight.

The 1930 record of the 92 apple orchards which were studied also in 1929 is as follows: 14 had no blight, 30 showed a trace, 22 had a slight amount, 11 had a moderate amount, and 18 were severely infected, varying from 10 per cent of the twigs or blossom clusters to the extreme of 95 per cent of the blossom clusters. In the 30 orchards studied in 1930 but which were not observed in 1929, one had no blight, two possessed a trace, five had a slight amount, two had a moderate amount, and 20 were severely blighted.

The total number of orchards examined in 1929 and 1930,

exclusive of duplicates, and the number of orchards showing a specified amount of blight is shown in Table 1.

TABLE 1. NUMBER OF ORCHARDS WITH SPECIFIED AMOUNTS OF BLIGHT IN 1929 AND 1930

Year	No blight	Trace	Slight	Medium	Severe	Total
1929	48	88	45	11	5	197
1930	12	32	26	12	40	112

In any orchard where the disease was present in amount more than a trace, the practice has been to count random lots of 100 twigs or flower clusters on various trees and record the number of blighted ones per 100. Thus, while one or a few trees in a given orchard may possess a considerable amount of blight, a count on representative trees in different parts of the orchard may reveal, as was often found to be the case, only a slight amount of blight. Conversely, when a part of an orchard showed little or no blight, the amount present on other trees would be such as to yield a relatively high figure for blight in the orchard as a whole. It may be noted, however, that the divisions of the amount of blight are merely relative and may or may not have any great significance with reference to losses in yield or in destruction of bearing wood. In those cases where blossom blight was very common or where twig blight involved more than 10 per cent of the branches, the losses in yields were considerable, often as much as 30 per cent on susceptible varieties in 1930.

The comparative amount of blight present in the same orchards in 1929 and 1930 is shown in Table 2.

TABLE 2. COMPARATIVE AMOUNT OF BLIGHT IN THE SAME ORCHARDS IN 1929 AND 1930

Amount of blight in 1929	Amount of blight in 1930 and the number of orchards involved in each amount				
	No blight	Trace	Slight	Medium	Severe
No blight (19) ¹ ---	4	2	6	3	4
Trace (43) -----	8	23	4	5	6
Slight (16) -----	1	4	5	2	4
Medium (9) -----	1	1	4	1	2
Severe (5) -----	0	0	3	0	2

¹Figures in parenthesis refer to the total number of orchards possessing a given amount of blight in 1929.

It is seen in Table 2 that out of 19 orchards which had no blight in 1929, 15 became infected in 1930, four of them, or over

20 per cent, severely blighting. And in the same sequence, out of 43 orchards which possessed only a trace of blight in 1929, 11 developed from 6 to 50 per cent of blight in 1930. It is very evident that there exists no positive correlation between the presence and absence of blight or in the amount of blight in 1929 compared with 1930 in these 92 orchards as a whole. It is only in specific instances that some correlation may be had between the current year's blight and that of the preceding year. In these instances particularly favorable factors for blight development, such as richness of soil, retentiveness of soil moisture or availability of soil moisture during certain parts of the growing season, favorable blooming periods in relation to inoculum dissemination, and other factors which are operative only in certain orchards, or parts of orchards, make for blight development year after year. The following field observations, made in conjunction with studies on the succession of blight, pertain to those instances where blight tended to be continuous: First, in certain orchards or portions of orchards which had formerly served as barnyards—in spite of the most arduous and painstaking efforts on the part of the grower to prevent the disease by pruning and other remedial measures; second, in parts of orchards which serve as poultry yards, in contrast to other parts in the same orchards which are separated from the poultry yards by means of wire fencing and in which blight is either entirely absent or present at times only in very slight amounts; and third, the more or less continuous appearance of blossom blight in portions of orchards which, on account of topographical differences, advance the blooming period several days ahead of nearby apples of the same variety, such advance usually making for blooming during bright sunny weather and hence for activity of pollinating and nectar-gathering insects.

Having cited instances in which blight is likely to occur in successive years, it seems desirable to cite specific cases where blight is occasional, and apparently unconnected with former blight in a particular orchard or in nearby orchards which may serve as sources of inoculum. The data in Table 2 show in a general way that numerous orchards which had little or no blight in 1929, suffered more or less severely from blight in 1930. In many instances these orchards were several miles removed from orchards or individual trees which might have served as sources of inoculum. A few typical illustrations may suffice. An 8-acre block of Yellow Transparent, located about 2 miles north of Springdale, Arkansas, owned by P. Brogdon, suffered such a severe infection from blossom blight in 1930 that about one-third of the crop was lost, with many of the trees showing 75 per cent of the blossom clusters blighted. This orchard had no blight in 1929 and a careful search for blight in the vicinity of this orchard in the same year revealed no cases where blight

was serious or present even in medium amounts. The Lucas orchard, about one-half mile away, showed a few scattered, blighted twigs; the Harp orchard showed a trace of blight; and the Riggs orchard also in the same vicinity, consisting of 16 acres of Jonathans, showed an occasional twig blighted in the orchard, with a few trees standing in a poultry yard having as much as 8 per cent blossom and twig blight. In 1930 not only the Brogdon Transparent orchard, but the Lucas Transparents as well were seriously blighted and a 3-acre portion of the Riggs' Jonathan orchard showed considerable twig blight, some trees having 60 per cent of the twigs blighted. Since the Brogdon orchard had no blight in 1929, it is obvious that no hold-over cankers existed in that orchard and that the severe blossom blight of 1930 must have been due to inoculum brought in from some outside source. Furthermore, the relative scarcity of blight within a 3-mile radius of this orchard in 1929, seemingly suggests that the possible activity of local hold-over cankers in exuding bacteria the following spring, and the dissemination of this exudate by rains, had little to do with the 1930 blossom blight epidemic in this vicinity. It will be seen later that as far as the United States Weather Bureau records show, no rain fell throughout the blooming period of apples either in this region or in any other locality in the apple growing section. As far as this whole group of orchards is concerned, there is evidently no correlation between 1930 blossom blight epidemic and the former year's blight, with reference to the initiation of the epidemic by exudates from hold-over cankers. Concerning blossom-visiting insects, the Brogdon orchard harbored a relatively large number of beehives which were located within a few feet from the block of Yellow Transparents.

Another illustration of the lack of succession or discontinuity of blight in a given orchard or in a group of neighboring orchards is exemplified in the I. M. Pound, C. C. Newham, Jack Weaver, and J. A. Eicher orchards north of Springdale, all within a radius of several miles. The combined apple acreage in these orchards totaled about 94 acres, each orchard containing among other varieties, Jonathan and Yellow Transparent. No blight was found in any of these in 1929. In 1930 the Pound orchard had a slight amount of blossom blight and about 20 per cent twig blight on Yellow Transparent, and a slightly greater amount of both kinds of blight on Jonathan. Other varieties, such as Ben Davis, Gano, Wilson June, Maiden Blush, Delicious, Early Harvest, and Stayman Winesap, had no blight. The Newham orchard showed 20 per cent twig blight and a trace of blossom blight on Yellow Transparent, a slight amount of both kinds on Jonathan, and no blight on Ben Davis, Winesap, and Gano. The Weaver orchard had a slight amount of twig blight on Yel-

low Transparent, while Jonathan showed a slight amount of both blossom and twig blight. Ben Davis, Winesap, Delicious, Maiden Blush, and Wilson June were not blighted. The Eicher orchard had a slight amount of twig blight on Yellow Transparent, while Jonathan had a slight amount of blossom blight and 20 per cent twig blight. Ben Davis, Maiden Blush, and Wilson June were not blighted. No blight having been found in these orchards in 1929, there was, therefore, no opportunity for local hold-over cankers acting as initiators of blight in 1930. Here also there was no succession of blight, and the most reasonable explanation is that the pathogen was brought in from an outside source. Numerous similar instances could be cited.

How do these findings compare with those in other states? In Wisconsin, Miller (20) found that primary blight in apple orchards is correlated with the presence of overwintered cankers and unpruned blighted twigs. Trees developing the greatest amount of blossom blight were the ones that had one or more unpruned, hold-over cankers or blighted twigs. On the other hand, other trees in the same orchard which had no overwintered cankers or blighted twigs showed little or no blight. In other words, his findings not only indicate a general relationship of current year's primary blight to that of the past year's blight in an orchard as a whole, but more specifically, they indicate a very direct connection on individual trees. Miller mentions no orchards where blight was absent one year and present in considerable quantity in the succeeding year. During the 3 years of his field observations, 1926 to 1928, "no general epidemic outbreaks of fire blight occurred" (*p. 617*) so that he dealt entirely with "local outbreaks." Unfortunately he limited his observations to a group of commercial orchards in one locality, Gays Mills, and evidently had little or no opportunity of observing any large number of blighted orchards in diverse localities. Thus, while his work on the succession of blight, which appears to the writer to be exceptionally thorough and painstaking, may be considered applicable to the orchards studied at Gays Mills in years when the disease is more or less localized and of minor importance as a whole, there is no proof presented that it would apply equally to the same section when the disease is widespread and of epidemic proportions, or to other apple growing sections possessing markedly different environmental conditions. This last point will be made clearer in the discussion on weather conditions in relation to blight development.

Contrasted with Miller's findings, Milbrath's observations in California⁴ on the severity and diffuseness of fire blight in 1930 compared with 1929 are quite in harmony with the writer's observations on the non-succession of blight in Arkansas. In

⁴Milbrath, D. G. Memorandum on blight in pears in 1930. 8 pp. (mimeographed) July, 1930.

1929 "less blight occurred in the state (California) than in a large number of years." In 1930, on the other hand "blight occurred on pear, apple and other hosts in all parts of the state. . . . There were instances of well isolated pear trees being severely attacked," and in a personal communication he adds, "How those trees became infected is quite a mystery. The general outbreak was almost simultaneous in all parts of the state about the third week in April." Evidently there was no correlation, in general, of blight in California in 1930 with that of 1929.

Pierstorff's observations in southern Ohio (*23*, p. 40) on the absence of active hold-over cankers in 1930, in orchards of Lawrence, Gallia, Meigs, and Athens counties, where severe epidemics of blossom blight had occurred, are also in harmony with the writer's observations. He states, "In most orchards no active hold-over cankers could be found, and yet up to approximately 90 per cent of the blossoms were blighted in extreme cases. No orchard was found entirely free from blossom blight. Comparable conditions were found on twenty-one farms in Brown and Adams counties"

From the foregoing discussion on the succession of blight, the following conclusions are drawn: First, the absence of blight in any year in a given orchard is no guarantee against the occurrence of the disease in the succeeding year. In seasons when blight assumes epidemic proportions, the disease will appear in orchards where blight was absent in the year previous, even though they are well isolated and out of reach of rain-driven inoculum; second, in those orchards or parts of orchards which are particularly susceptible to blight, because of local environmental conditions, the disease is likely to appear in successive years in spite of arduous efforts to prevent it by pruning operations.

THE RELATIONSHIP OF WEATHER CONDITIONS TO THE DEVELOPMENT OF BLIGHT

Within recent years much valuable information has been presented on the relationship of weather conditions, particularly rainfall, to the development of fire blight. Since this has been reviewed very recently by Pierstorff (*23*) there is no need of going into details. Suffice to say that careful workers in Wisconsin (*2, 20*) and Michigan (*41*) have found that rains falling over exuding, hold-over blight distribute the bacteria to susceptible organs located below the overwintered blight or in the path of bacteria-laden raindrops. Tullis (*41*), Miller (*20*), and the writer (*29*) have also found that in the presence of water *Er. amylovora* may penetrate young leaves; likewise petals, sepals, and outer walls of receptacles may serve as infection courts (*29*), and that

such penetration may occur in the absence of wounds or insects of any kind. Miller and Tullis have, therefore, concluded that the initial dissemination of blight is due to rain falling on bacterial exudates engendered in hold-over blight and washing the bacteria upon susceptible tissues. They present weather records tending to show a correlation between rainfall and blight development. Pierstorff (23), working in Ohio and New York, however, was unable to correlate rainfall with blight development, and since he failed to produce foliar infections in the absence of insects, he concluded that meteoric water plays little or no part in the spread of the disease.

In considering the role of weather conditions in relation to blight development, it may be helpful to call attention to the fact that fire blight is not restricted to those portions of America which experience rains, dews, or mists during the growing season. Indeed, throughout extensive areas in western America where irrigation is necessary for crop production and where meteoric water is of rare occurrence in periods of active growth, fire blight is often present and occasionally very destructive. The severe epidemics of blight on pears in the arid, interior valleys of California are too well known to require comment. Obviously in some portions of America there seems to be little or no connection between meteoric water and the occurrence of fire blight, although this requires further work for its substantiation.

Fire blight usually makes its first annual appearance in Arkansas shortly after the first petal fall, in the latter half of April, Table 3 and (29, p. 16). Hence a study of weather condi-

TABLE 3

	Host	Flowering period	Frost injury to blossoms	Date of first appearance of blight	Relative prevalence of blight during season
1930	Kieffer....	March 3 to Apr. 1	99% killed ¹	April 11	Very slight
	Jonathan...	Apr. 5 to Apr. 12	10% killed ¹	April 14	Extremely severe
1931	Kieffer....	March 14 to Apr. 5	95% killed ²	May 4	Slight
	Jonathan...	Apr. 7 to Apr. 21	30% killed ²	May 4	Slight to moderate
1932	Kieffer....	March 3 ³	100% killed	April 27	Very slight to absent
	Jonathan...	Apr. 8 to Apr. 20	absent	April 25	Slight to moderate

¹The severe January freeze of 1930 injured the buds and bearing wood of pears to such an extent that on most trees all the blossoms dropped off. On apples this freeze injured the wood to a considerable extent, in some localities the bearing wood being so severely injured that many of the blossoms failed to set fruit.

²On March 27, 1931, a severe freeze occurred when most of the pear trees were in full bloom and when many of the apple buds were beginning to separate in the cluster.

³On March 5, 1932, and extending to March 9, a severe freeze occurred, killing all pear blossoms just as they were entering full bloom.

tions prevalent in the fruit-growing section during the early part of the growing season may throw some light on blight development.

On Kieffer pears, early growth including flower production often occurs in periods of frost, or intermittent periods of cold and warm weather, frequently resulting in the death of a large portion of the blossoms (Table 3). Indeed both pear and apple blossoms are commonly produced in seasons of extremely fluctuating temperatures, with cool, often frosty, nights and warm days. This often results in relatively long drawn out flowering periods, with the early blossoms opening two to three weeks prior to the later ones on the same tree. In contrast with the published records (20) of early growth and flowering of apples in southern Wisconsin, which show relatively short blossoming periods of about 9 days to 2 weeks, Arkansas apples, with relatively rare exceptions, take about twice as long to bloom. (Figures 4 and 5.) When blooming periods are thus prolonged, a variety of weather conditions is very likely to be encompassed. The early portion of blossom development may occur during a period of warm, sunny weather, which is succeeded by very cool, wet weather, which in turn is replaced by a warm interval, wet or dry, and so forth. The flowering periods of 1927 and 1928, shown in Figures 4 and 5 were typical examples of such irregularities.

Such prolonged blooming periods taking place during diverse weather conditions make it exceedingly difficult to evaluate the influence of some particular environmental factor on the spread and development of the disease, and help explain why some observers have correlated early blight development with cool, wet weather; others with warm, wet weather; and still others with dry, warm weather. Since all of these conditions may be present in any one flowering period, the observer need but choose those which he thinks are influential in blight development, and disregard the others. No exact information being at hand on the influence of any particular combination of weather conditions on blight development, one has a relatively wide choice. A more exact picture may be had of these difficulties by following a detailed account of weather conditions existing during the blooming season of apples at Fayetteville in 1927.

On March 17, 1927, a mild sunny day, Jonathan flower buds were separating in the clusters, following a heavy rain the night previous. The next two days were rainy and equally as mild. Then came a 3-day period of frosts and rain, with the temperature going as low at 26 F., resulting in some injury in the form of brownish discoloration to petal tips and to a small number of receptacles, in some injury to leaf buds which became blackish

and watersoaked, and in killing fully 95 per cent of Kieffer pear blossoms. This was followed by bright but cool weather for 5 days in which Jonathan blossoms continued a slow development, the petals attaining a full pink color and the pedicels developing to about two-thirds of their full length. Then came a 4-day rainy period, cool at first and gradually becoming warmer, the Jonathan flower buds attaining full size. The next day, April 1, the first Jonathan blossoms opened, on a cool, windy and sunny day. Following another cool, bright day, with but little growth, the weather became quite warm and continued bright, with about one-third of the blossoms opening on April 3. The next day was again unseasonably warm, and sunny, with about 60

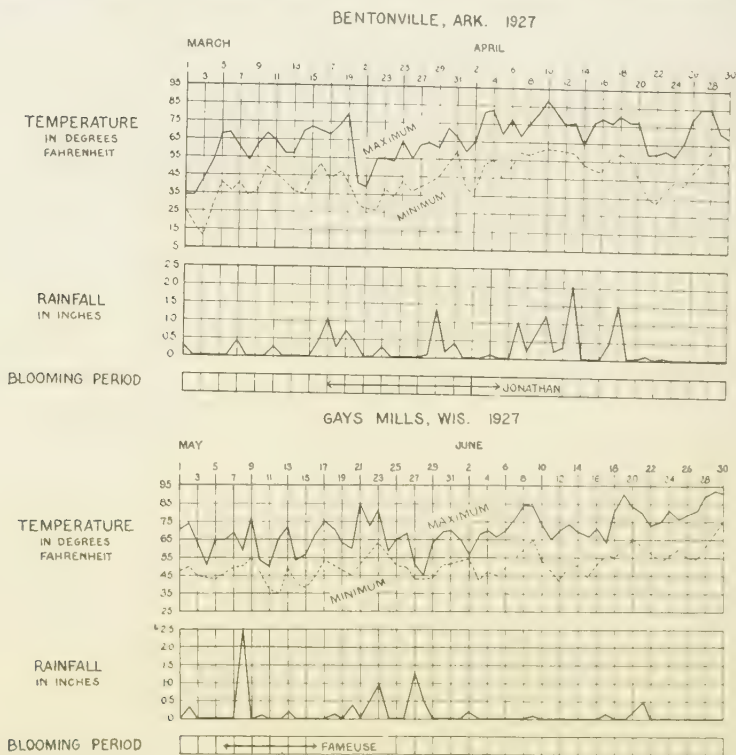


Figure 4. A comparison between weather conditions at Bentonville, Ark., an important apple growing area, and Gays Mills, Wis., during early growth and flowering of susceptible apple varieties in 1927. The data for Bentonville are taken from U. S. Weather Bureau records, while those for Gays Mills are from Miller (21).

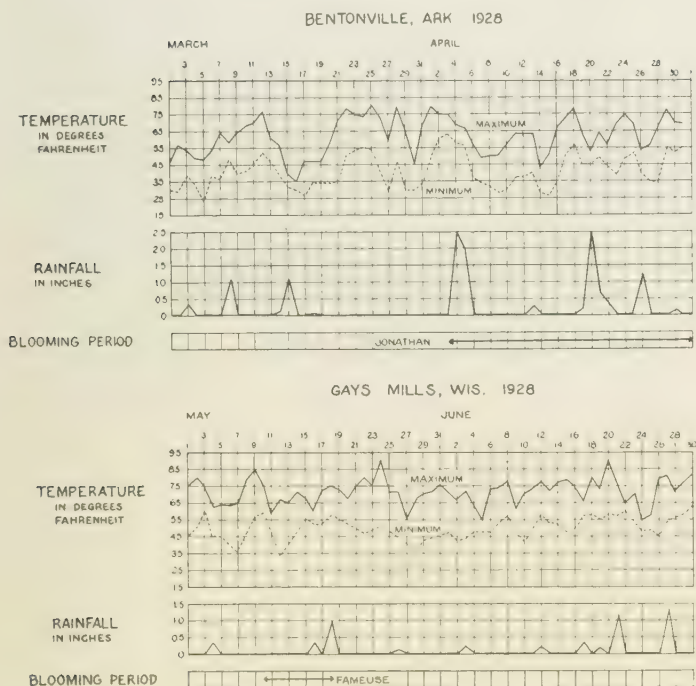


Figure 5. A comparison between weather conditions at Bentonville, Ark., and Gays Mills, Wis., during early growth and flowering of susceptible apple varieties in 1928. The data for Bentonville are taken from U. S. Weather Bureau records, while those for Gays Mills are from Miller (20).

per cent of the flowers opening to full size. On April 5, another bright but cooler day, 90 per cent of the blossoms were fully open and by April 7 in a continued period of sunny weather, many of the Jonathan fruits had apparently set. Throughout the period of opening of blossoms no rain fell at Fayetteville, while at Bentonville a slight rain of 0.1 inch fell on April 4, the remainder consisting of warm, sunny weather as at Fayetteville. From April 7 to April 24 rain fell almost daily, with warm weather up to the twentieth, and a sudden cold snap on the twenty-first and twenty-second, culminating in a heavy frost on the morning of the latter day. Fortunately the young Jonathan fruit had grown sufficiently that it escaped any considerable amount of injury.

The first blight, in the form of a large number of dead Jona-

than fruit clusters, was detected on April 15. Microscopic examinations of the diseased tissues revealed great numbers of typical fire-blight bacteria. No pure-culture isolations were attempted. On the next day more fruit clusters and a few leaf clusters on Jonathan were found blighted. As artificial inoculations conducted at this period with pure cultures of *Er. amylovora* in water suspensions, atomized on open apple blossoms in the orchard, produced no signs of infection until 10 to 12 days after inoculation, it would appear that the natural infections noted on April 15 and 16, were probably initiated on April 3 to April 6. This period was characterized by warm, sunny weather in which bees and other pollinating insects were very active. It would thus appear that such insects may have been the agents involved in disseminating the bacteria. Nevertheless, despite the correlation of rainless weather with opening of blossoms, there is in fact no good evidence which would entirely exclude rain as the disseminating agent. Rain having fallen both before the blossoms opened and shortly after petal fall, there is the possibility that dissemination was brought about by meteoric water, and that infections took place not by way of floral nectaries but on either leaf tissues located on fruit spurs or on some portion of the blossom other than the nectarial surface. The writer has shown (29, p. 54) that pear flowers may become infected by way of peduncles, receptacle walls, calyx lobes, and petals; and, although Rosen failed to produce infections on apple blossoms through similar tissues, Pierstorff (23, pp. 31-32) succeeded in producing infections through apple petals and stigmas, without wounding. However, the latter author concluded that under normal conditions apple blossoms are not likely to become infected in this manner.

Assuming that pollinating insects were responsible for the dissemination of blossom blight in 1927, how does one account for the presence of blight on leaf clusters, noted on April 16, which were not associated with blossom clusters? Evidently to attempt to correlate blight development with either insects or rainfall in 1927 in Arkansas or in any other section of America where rains alternate with sunny weather and pollinating insects are present in abundance, is fraught with considerable difficulty. In Arkansas, the problem of early spring disseminating agents is further complicated by the fact that during a 6-year period, including 1927, no bacterial exudate from overwintered blight has been found prior to the initiation of spring blight, as previously detailed. As far as the initiation of blight in 1927 is concerned, instead of excluding either rainfall or insects, the evidence suggests that both of these may have played a part as disseminating agents.

If more conclusive information were desired on the role of

either insects or rains as early disseminating agents, it is clear that some season or location must be studied in which either rainfall or insects is absent throughout the early period of growth. In this connection, the year 1930 in Arkansas offers a highly interesting period (Figure 6.) This was the notorious year of severe drouth, and also the year when fire blight on apples in Arkansas and pears in the Pacific states, to mention but two localities, suffered from one of the worst epidemics of blight that the country has ever experienced.

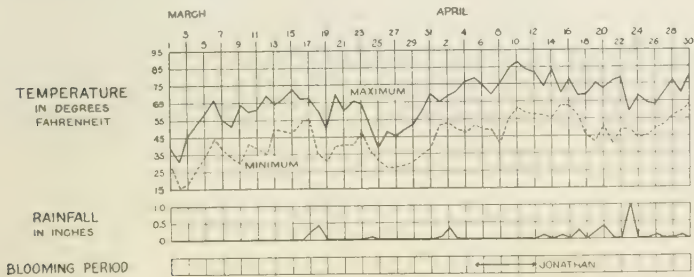


Figure 6. Weather conditions at Bentonville, Ark., in 1930, during early growth and flowering of Jonathan. (U. S. Weather Bureau records.)

In 1930, early growth and flowering of Jonathans, as well as other susceptible varieties of apples, began in the first week in April at both Bentonville and Fayetteville. The previous month had been exceedingly dry, 0.82 inches of rainfall being recorded for the whole month at Bentonville. The first 3 days of April, as the Jonathan flower buds were beginning to swell, marked a warm and slightly rainy period, the total rainfall for the 3 days being 0.41 inches. This was followed by a 9-day period of unseasonably warm, dry, and sunny weather, during which apple-blossom development, from the early cluster-bud stage to the setting of fruit, was encompassed. Such telescoping of practically the whole blossoming period into a period as short as 9 days, constituted one of the outstanding phenomena experienced in this most exceptional year. Very little rain fell during the remainder of the month so that the United States Weather Bureau reported that "this was the driest April in Arkansas since state-wide records have been kept."⁵

The first blight on apples was noted at the end of the second week in April, in the form of blossom blight, and by the end of the third week, blossom blight had assumed such widespread proportions that it was difficult to find any fair-sized, bearing

⁵U. S. Department of Agriculture Weather Bureau. Climatological Data—Arkansas Section, April, 1930.

orchard of susceptible varieties that had escaped. As previously noted, it was found in numerous orchards which had suffered no blight in 1929. Without exception blight was found only on trees that had produced blossoms, and was at first entirely absent on other trees which, because of age, varietal behavior, or adverse circumstances, had not bloomed. Many cases were noted in which young, non-blooming resets standing adjacent to older trees which had borne blossoms, had completely escaped infections when the older, bearing trees showed a large amount of disease. The first twig blight on apple was noted on April 22, about one week after blossom blight had been found. It was observed in an orchard at Farmington, where blight was present in the previous year. Here it seems desirable to call attention to the observations recorded by Brooks (2) and Miller (20) in Wisconsin in which newly formed apple shoots or twigs were found blighted prior to the appearance of the disease on blossoms. Now in order to get twig blight, the twig must first be produced, unless it is assumed that the infection occurred in the bud stage, an assumption which is not postulated by these writers. Twig growth in Arkansas on all common varieties of pears and apples grown in this state, does not ordinarily appear in noticeable amount until petal fall, so that when blossom blight appears, it usually is observable about a week or two before twig blight. Indeed, it is not at all difficult to find blossom blight present on trees when twig blight is entirely absent, although these trees may later show twig blight. In Arkansas the first form of blight to appear on apples is usually blossom blight, as it was in 1930.

No rains having fallen throughout the whole period of blossom development on apples, it is clearly out of the question to attempt to correlate the severe epidemic of blossom blight in 1930 with rainfall. Rain evidently could not have acted as a disseminating agent on blossoms in this season. On the other hand, one may reasonably assume that the exceptional warm, dry weather, having forced an extraordinarily rapid floral growth, had increased the susceptibility of the floral parts to disease. This is in harmony with numerous observations on fire blight in which investigators over a long period of years have found that rapidity of growth makes for blight susceptibility, the disease being almost entirely restricted to those portions of the growing season when growth is most rapid and being as a rule inconsequential in the latter half of the growing season when growth has slackened.

The warm dry weather during the blooming period of apples also made conditions ideal for the activity of pollinating and nectar-seeking insects. It is well known that bees are not active during periods of rain or of cool or windy weather. The warm,

sunny weather continuing day after day enhanced the activity of bees and other insects to a considerable degree. This activity coupled with the fact that during the previous winter and at various times in the early spring of this season, the writer succeeded in isolating the fire-blight pathogen from beehives located in apple orchards, lends considerable weight to the idea that blossom-visiting insects were primarily responsible for the dissemination of blossom blight on apples in 1930.

The history of fire blight on pears in Arkansas in this season is quite different from that of apples. The severe January freeze of 1930 resulted in extreme trunk and limb injury to apples as well as in a complete killing of pear blossoms. On all pear trees under observation, the blossoms dropped before any signs of blight were discernible. The first blight symptoms on pear were found at Bentonville on April 11, 3 days ahead of the finding of blossom blight on apples. Six different infections on Kieffer pears were noted, five of which were twig blight. The low-temperature injury to blossoms, which resulted in the death of ovarial tissues, although apparently not interfering with other floral organs, had forced an early and rapid twig growth. As twig blight cannot easily be traced to the activity of pollen-seeking insects, it is obvious that they cannot be held responsible for the dissemination of the disease on these pear twigs.

On the other hand, the finding of blight on pear twigs only 3 days in advance of the detection of apple blossom-blight does not necessarily indicate any relationship in source of inoculum between pear and apple or in kind of disseminating agent operating on apple compared with pear. Had not the pear blossoms been injured by the freeze, there is very good reason to believe that some of these blossoms would have developed blight either prior to the appearance of twig blight or at least at the same time. Kieffer pears were in full bloom from March 28 to March 31, 11 to 14 days preceding the appearance of pear twig blight. It will be shown later that fire-blight bacteria may be present in pome flower tissues, but if some adverse condition such as frost or winter freeze prevents the fruit from setting, the blossoms may be dropped irrespective of the presence of the pathogen within the tissues. Thus an infection which may have originated in a blossom and have extended into a twig which arose from the same fruit spur might readily be mistaken for primary twig blight. If the blossom infection has not advanced downward sufficiently to involve twig or spur tissue, the bacteria multiplying in the floral nectaries, a well authenticated phenomenon (42, 44, 6), or exuding from floral organs, may still be a source of inoculum for later twig blight or for later blooming varieties despite the insecurity of that particular blossom. Nevertheless the finding of twig blight on pears in the early part of the grow-

ing season, 3 days ahead of its detection on apple blossoms, still points to the possibility that some agent other than pollinating insects may have been involved in disseminating the pathogen on pears at that particular time, and that this agent was either in the form of rain, (the light rainfall of April 1, 2, and 3), or some insect other than bees. As a working hypothesis the writer at present is accepting the former agent rather than the latter for reasons that will be apparent later.

Following the very dry warm April, May, 1930, was unseasonably cool and exceptionally rainy, rainfall being above normal in all portions of the apple region, except at Rogers. Until May 18, rain was almost of daily occurrence around Springdale, Fayetteville, and Farmington, in Washington County, and less common at Bentonville and at Rogers in Benton County. All of these localities are important apple growing centers. The remainder of the month, with the exception of May 23, consisted of dry sunny weather, ushering in a drouth which lasted throughout most of the growing season. While a slight amount of twig blight on apples had been observed as early as April 22, in most orchards twig blight was either wholly absent or present only as a trace until the third week in May. Then appeared a very severe outbreak of twig blight, particularly in orchards around Farmington and Springdale where rainfall had been heavy. At Bentonville and Rogers, which had experienced less rain, twig blight was not nearly as severe. By May 31, it had involved so much of the periphery or tops of trees in some orchards that viewed from a distance, they appeared to have been struck by some widespread and all-consuming conflagration. In the 15 years of plant disease study in Arkansas, the writer has never seen such destructiveness from this or any other disease.

The amount of twig blight in numerous orchards did not show any direct relationship to the amount of blossom blight that had developed earlier in the season. In some orchards which had suffered severely from blossom blight, twig blight which was unconnected with infected blossoms was rare. In other orchards all the trees of one variety or only a group of trees with much blossom blight and little twig blight were observed standing near other trees of the same or different variety which had very little blossom blight and a large amount of twig blight. Lastly, twig blight was found in orchards where no blossom blight was detected. This, however, was relatively rare and was mostly confined to two kinds of orchards, to those in which blight had been present in a greater or less degree in 1929, and to those in which, because of severe winter injury, there had been a very light set of fruit. In the last instance the number of leafy shoots and size of individual shoots were seemingly increased by the scarcity of fruit production, and it is quite

likely that this stimulus for vegetative growth made for increased susceptibility of twigs.

Recapitulating, field studies of 1930 indicate that the main cycle of apple blossom blight in that year was not associated with rainfall but with the activity of pollinating insects, while the chief outbreak of twig blight was associated with rainfall and not with insects. During the 6 years in which careful records of weather conditions and insect activity in relation to blight have been kept, no year offered greater contrasts as between dry and wet weather and as between presence or absence of insect activity than the season of 1930. It would burden this account to present the records of weather conditions and insect activity during 1929, 1931, and 1932. In these years no such clear cut correlation was obtainable as in 1930. Furthermore these years as well as 1927 and 1928 saw no such widespread epidemic of fire blight in Arkansas as 1930.

SEQUENCE OF FIRE BLIGHT WITHIN A GROWING SEASON

FIRST BLIGHT ON PEARS

A study of the sequence of blight during the growing season, with special reference to the particular organs and tissues that blight at different times, is of considerable interest. If it is found that blight is restricted to certain parts of the trees or to certain organs at one time of the growing season and to other parts at other times, it is possible that valuable clues may be had as to the origin of inoculum, agents of dissemination, and possible control measures.

In a previous article (30) the writer has noted that, with relatively rare exceptions, the first signs of blight in bearing orchards are to be found in the blossom clusters. While this had reference primarily to Arkansas conditions, there are numerous notes by other investigators in other states which present the same finding. Among these are Waite (44, 45, 46, 47, and 48) the locality not being designated, Whetzel and Stewart (50) in New York, Sackett (34) in Colorado, Stewart (38) in New York, Gossard and Walton (6) in Ohio, Hesler (11) in Tennessee, Owens (22) in Oregon, Anderson⁶ in Illinois, and Cunningham (4) in New Zealand. Likewise, the findings of numerous other investigators are seemingly in harmony with this view, although their accounts are not sufficiently specific on this point. Many make the statement or imply that both blossoms and leafy shoots show the disease more or less at the same time in the early part of the growing season. According to the writer's knowledge, there are only two investigators, Brooks (2) and Miller (20), who specify that leafy shoots are the first to exhibit symptoms

⁶Plant Disease Reporter, U. S. Department of Agriculture Bureau of Plant Industry 13:16, 1929.

of blight. It is evident that if in any section of the country the first blight of the season is restricted to blossom clusters and is not present on leafy shoots until later in the season, then some factor must be considered as the disseminating agent which is active only on floral parts. Unless it is assumed that blossoms are much more highly susceptible than young, partly embryonic leaves, an improper assumption, rain and wind may perhaps not be expected to play a role, since they would affect leafy shoots as well as flower clusters.



Figure 7. Artificial infections on Kieffer blossom clusters inoculated with a pure culture of *Er. amylovora* in water-suspension, applied as a spray which simulated rainfall. Note infections on sepals, receptacles, pedicels, and tips of leaves. Photographed 3 days after inoculation.

As early growth and flowering appears first on pears (29), this discussion will concern pears first and apples later. In Arkansas, when pear blossoms are wide open, leaves both in the flower clusters and from leaf buds have attained a size of about 1 to 2 cm. in length and have partly uncurled from the bud stage (see Figure 7). At this time there are very few or no leafy shoots observable. Leafy shoots do not become evident on Keifer pear until petal fall or thereafter, when sufficient stem

growth has developed so as to be designated a shoot. Such shoots often arise first from the flower clusters or fruit spurs, especially when frost or low temperatures have injured the blossoms, and are frequently 2 to 4 cm. in length before the terminal or lateral leaf buds have grown sufficiently to be designated as leaf shoots. Thus when Kieffer pear blossoms have separated in the cluster, there are numerous leaves beginning to unroll, both in the flower clusters and in the strictly vegetative buds, but ordinarily there are no shoots as yet. By applying a water spray of a pure culture of *Er. amylovora* to pear branches which are at this stage of development, many infections can be induced on both leaves and floral tissues, and the relative susceptibility of leaves and flowers at that stage of growth may be determined. As some of the writer's work on blossom infections has been discussed in a previous publication (29) only sufficient data will be presented here to clarify the critical points.

In order to determine the relative susceptibility of floral tissues as contrasted with leaf tissues, numerous inoculations were attempted both under natural field conditions and in the greenhouse, involving several thousand blossoms. Pear blossoms and leaves, being much less hairy than most apple varieties, offer much superior material for study of susceptibility, since the infection centers, period of incubation, and rate of disease spread are far more easily determined on a slightly hairy surface than on one obscured by a heavy pubescence. Early in the work it was found that artificial infections in the field were not always obtainable by methods that invariably produced infections in the greenhouse. Lack of control of various environmental factors being difficult and sometimes impossible under natural conditions makes field studies of this nature somewhat precarious and uncertain. However, as far as the crucial points are concerned, artificial inoculations made in the orchard fully confirm the greenhouse tests. These investigations revealed the following:

First, at the time of blossom production, young leaves located in the blossom cluster or arising from vegetative buds, are fully as susceptible to infection as any floral organ. Second, infections on flowers and leaves are readily obtainable in the absence of insects or wounds by the application of water-borne inoculum applied as a watery spray.

As a typical example of such findings, the following experiment is detailed. On January 21, 1933, a number of Kieffer limbs bearing swelling buds were severed from trees, the bases of the limbs being immersed in water in 2-gallon jars, and kept in the greenhouse until January 27. On this date many of the individual flower buds were separated in the cluster, the calyx lobes in most instances firmly and completely enclosing the other floral parts. Only a few leaves were sufficiently advanced in growth

to be distinguished in the blossom cluster. Very few petals were exposed. The control, treated with a spray of sterile water applied by a sterilized, all-metallic artist's atomizer, consisted of 63 blossoms comprising 11 separate clusters. A 2-weeks old broth culture of *Er. amylovora*, diluted one to one with sterile water, was similarly applied to 111 blossoms comprising 18 clusters. The controls as well as those opening buds inoculated with the pathogen were immediately placed in a cloth-enclosed moist chamber, possessing an overhead watering system so that a fine mist was maintained. The temperature of the greenhouse throughout the experiment varied from about 20° to 30° C., the average or mean temperature being close to 25° C. (77° F.). Within the moist chamber the temperature was probably slightly lower. The mist was permitted to fall for 24 hours and then discontinued. The limbs immersed in water were kept in the chamber for 72 hours, and then placed on an open greenhouse bench. At the end of this period, on January 30, the following notes were taken: Of the inoculated branches, there were 135 sepal infections, 13 infections on receptacle walls, 24 pedicel infections, 8 spreading infections, involving sepals, receptacles, and pedicels, 4 petal infections, and 12 leaf infections, a total of 196 separate infections. The controls showed no infections. In each instance the infection had developed sufficiently within the 72 hours to reveal clear symptoms of discoloration (see Figure 7) often accompanied by droplets of exudate. Microscopic examinations invariably revealed great numbers of bacteria within the infected areas (see Figure 8).

Practically every leaf exposed to the inoculum became infected, the infections in most instances starting at or near the apex (Figures 7 and 9), and often involving considerable laminar tissue along the midrib. A microscopic examination of these leaves revealed numerous stomata on the dorsal surface near the apex, with excellent indications of penetration having occurred through these stomata, masses of bacteria being clearly observable in substomatal chambers and in adjoining air spaces. No insects were found on any of the limbs, flower clusters, or leaves.

Numerous similar experiments conducted during the last few years have yielded comparable results (see Figure 9). There can be no question whatever that, given suitable environmental conditions, a watery spray of fire-blight bacteria simulating wind-driven raindrops laden with bacteria or simulating merely a falling of bacteria-bearing raindrops, is sufficient to produce infections on both young leaves and floral organs. On older leaves infections under similar conditions are either rare or absent, and it is probable that Pierstorff's failure (23, p. 37) to obtain leaf infections in the absence of wounds or insects may have been due to the use of plants which were lacking in young, part-

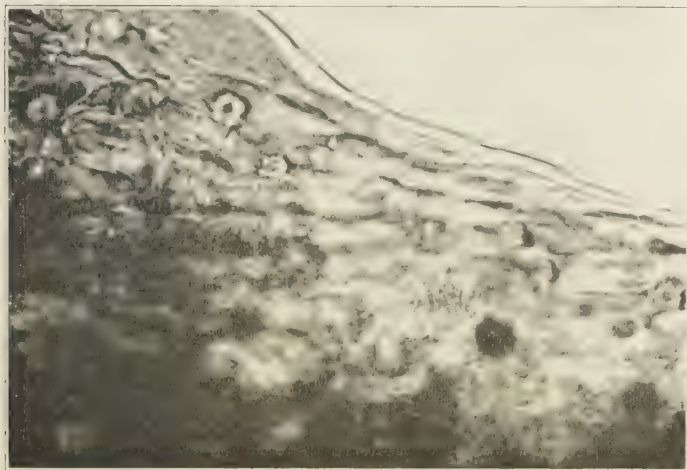


Figure 8. One of the artificially infected, Kieffer calyx lobes shown in Figure 7, depicting great numbers of bacteria within the infected area. Lobe immersed in Amann's mounting fluid containing cotton blue and heated slightly. Magnified 690 X.

ly embryonic leaves. In other words, these artificial inoculation experiments clearly suggest that the young leaves present at the time of blossom production are as susceptible as floral tissues to invasions by water-borne inoculum, and that in the presence of such inoculum, both leaves and flowers become subject to infection.

Under natural conditions, are the first infections to be found on both leaves and flowers, independent of each other, as they are by application of water-borne pure culture inoculum? It is to be observed in Table 4 that in only one year out of six was there no frost or cold weather injury to pear blossoms. This means that this one year, 1929, is the only one in which first signs of blight were not complicated by frost injury to blossoms, a type of injury which is very likely to obscure blossom infections. Not only are the symptoms of frost injury very similar to those of fire blight, but as previously noted, there is excellent evidence to show that infections may occur in blossoms resulting in considerable tissue invasion, and when frost strikes these blossoms there will often be no noticeable external symptoms of disease, aside from the occasional presence of very inconspicuous droplets of bacterial exudate. Finally, the frosted blossoms may drop before the disease is likely to be detected in them.

The evidence for blossom infections occurring in the ab-



Figure 9. Artificial infections on Garber pear blossom clusters, inoculated with a pure culture of *Er. amylovora* in water-suspension, applied as a spray. Note numerous infections at leaf tips as well as on blossoms. Compare with one of the control blossom clusters shown in the lower part of the photograph.

sence of disease symptoms was obtained on May 4, 1931. At that time certain Jonathan flower clusters which had failed to set fruit and which evidently had been seriously injured in the bud stage by the March 27-28 freeze, were noted with minute droplets of ooze adhering to some of the receptacle walls and tips of calyx lobes. No signs of leaf infections were observed. These blossoms had abscised from the spurs and were about to drop. No reddish-brown discolorations, typical of fire-blight symptoms of apple flower and leaf, were observed, and even the droplets of exudate were not by themselves sure signs of blight. The writer has frequently found droplets of ooze at the tips of calyx lobes, at tips of leaf scales and bracts, at tips and margins of young pear and apple leaves, and at places which had suffered from insect or mechanical injury, which upon microscopic examination revealed at times bacteria which proved to be non-infectious when injected in water suspensions into healthy, succulent pear sprouts. Such droplets are common, particularly in the early part of the growing season and are doubtless due either to a guttation phenomenon occurring at apical and marginal hydathodes or to release of water droplets at wounds as a response to the severing of water ducts. Apical hydathodes of calyx lobes are relatively large and readily observable, particularly on pears, by mounting a lobe in Amann's fluid and heating. The clearing reveals a spacious pore at the tip of the lobe, with vascular strands leading directly to the pore (see Figure 10). On these calyx lobes are also found large glandular hairs, located at the margin.

When some of these droplets of exudate observed on blossoms which showed no blight symptoms were mounted in water in a hanging-drop and subjected to microscopic examination, they were found to be thickly populated with bacteria of a type very comparable to *Er. amylovora* (see Figure 11). Likewise the tissues to which the ooze had clung, when examined similarly, were found to contain enormous numbers of bacteria. One of these blossoms was surface sterilized with mercuric chloride, washed in sterile water, mashed in a sterile Petri dish containing about 10 cc. of sterile water and used for inoculations into 3 healthy pear shoots by means of a sterilized hypodermic syringe. Within 3 days all the inoculated shoots developed typical blight symptoms when uninoculated trees standing on the same greenhouse bench remained free from disease. From one of these shoots the organism was recovered and single colony isolations were obtained from a series of poured plates. A transfer made from one of these colonies, when diluted with sterile water and injected into healthy pear shoots, again produced fire blight.

There can be no doubt that these frost injured, abscised blossoms had been infected with fire blight, although they show-



Figure 10. Apical hydathode on calyx lobe of Kieffer pear. Material immersed in Amann's mounting fluid containing cotton blue and heated slightly. Magnified 560 \times .

ed no outward signs of blight as far as discoloration of tissues was concerned. Again, the production of ooze clearly indicates not only that the disease had been contracted a number of days prior to the finding of these infected blossoms, but that such ooze might readily have served as inoculum for secondary infections on leafy shoots.

Thus the presence of a slight amount of twig blight on both apple and pear found at the same time as these infected blossoms (Table 4) cannot with certainty be ascribed to the same original inoculum, whatever its source may have been, or to the

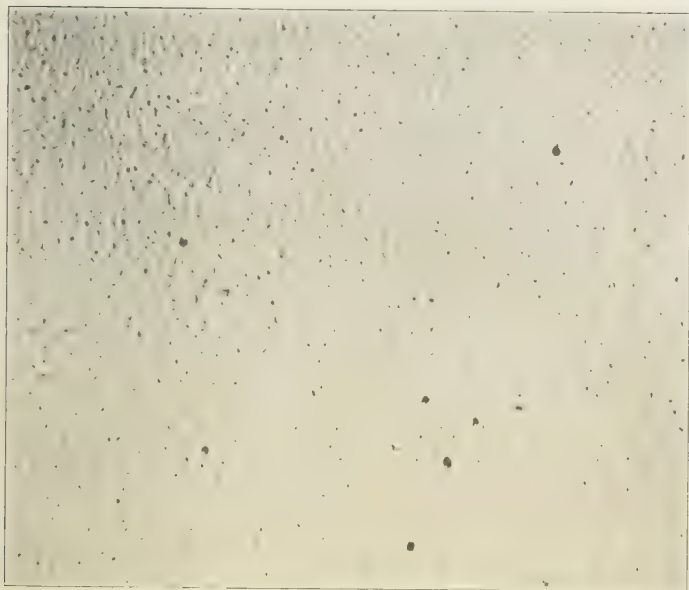


Figure 11. Unstained, water-mount of a droplet of dried exudate which clung to a pedicel of a Jonathan flower. This flower together with all others in the cluster had abscised from the spur, and when observed was about to drop. It showed no outward symptoms of blight, although the pedicel when surface sterilized, mashed in sterile water, and used for a series of dilution plates yielded pure cultures of *Er. amylovora*. Single colony transfers produced typical blight symptoms on healthy pear shoots. Magnified 530 \times .

same disseminating agent. Indeed, as most of the blight on pear and apple found at that time was blossom blight, with very little twig blight and no leaf blight which was independent of blossom blight, it seems more reasonable to assume that blossom blight appeared ahead of twig blight. This theory is in part substantiated by the fact that the main outbreak of twig blight in 1931 was not observed until May 28 on pears, and not until July 6 on apples. The few leafy shoots on pears and apples found blighted on May 4, when many blossom clusters were found blighted on apples, and when most of the pear blossoms had dropped because of the March freeze, can more reasonably be considered as secondary infections than as primary ones. When it is recalled that in this region there is no new shoot-growth on pear at the time of blossoming, which in 1931 was from March 14 to April 5, and when it is further recalled that no leaf infections were found that were independent of flower infections, it seems

TABLE 4. DATES OF DETECTION OF BLOSSOM AND TWIG BLIGHT ON PEARS AND APPLES, 1927-1932

		1927	1928	1929	1930	1931	1932
Pear	blossom blight---	01 (?)	02 (?)	Apr. 10	Apr. 11 ⁴	May 4 ⁵	06 (?)
	twig blight— first infections---	Apr. 16	Apr. 23	Apr. 25	Apr. 11	May 4	Apr. 27
	twig blight— chief outbreak---					May 28	
	twig blight— last infections---	July 12	July 31			Sept. 8	May 2
Apple	blossom blight---	Apr. 15	Apr. 29	Apr. 16	Apr. 14	May 4 ⁵	Apr. 25
	twig blight— first infections---	Apr. 18	May 3	Apr. 25 ³	Apr. 22	May 4	Apr. 25 ⁷
	twig blight— chief outbreak---	May 6	June 21	June 15	May 26	July 6	June 13
	twig blight— last infections---		June 28	July 19	Aug. 15	July 10	

¹Nearly all pear blossoms killed by frost.

²Fully 50% of the pear blossoms were killed by frost, some trees losing all their blooms. The particular trees under observation for fire blight lost all their blossoms from frost injury. This was so severe that in addition to the killed blossoms, many leafy shoots were injured, with symptoms so comparable to fire blight that inoculations had to be made in order to ascertain the nature of the injuries. Out of 33 artificial inoculations with blackened twig tissues, macerated in sterile water, 30 produced typical fire-blight infections on healthy pear trees maintained in a greenhouse.

³The twig blight found on April 25 was in all instances directly associated with blossom blight. The chief outbreak of twig blight did not appear until June.

⁴Complete killing of ovarian tissues with no fruit set, as a result of January freeze.

⁵Freeze of March 27 killed fully 95% of the pear blossoms and many apple blossoms.

⁶The freeze of March 5 to 9 resulted in the death of all pear blossoms and in some injury to apple blossoms.

⁷One leaf-shoot found blighted which was not connected with a flower cluster. It was located 2 inches below a blighted flower cluster and when the tissues connecting the two were examined under a microscope, bacteria were found throughout the whole length of this connecting tissue.

more reasonable to assume that blossom blight occurred first and that the few leafy shoots found blighted on May 4 represented secondary infections. (The origin of shoot or twig blight will be discussed later.) The March freeze having killed most pear ovaries and some apple ovaries, and having considerably retarded the growth of those which managed to live, probably prevented the external expression of disease symptoms on the blossoms as early as they otherwise may be expected to appear. To the writer this appears at present to be the best explanation for the finding of much blossom blight and a very slight amount of twig blight in the early part of the growing seasons of 1930, as well as 1931. It also explains the absence of blossom blight on pears for the years 1927, 1928, and 1932, when practically all such blossoms were killed by low temperatures.

This explanation for the finding of both blossom and twig blight on pears in the early part of the growing season is further substantiated by the record of 1929, when low temperatures played no part in floral injury. In this year the earliest blight was



Figure 12. Natural fire-blight infections on pear blossoms, the earliest infections of the season. Photographed April 16, 1929. Note absence of blight symptoms on leaves and on lower portions of flower stalks, indicating the visitation of contaminated, nectar-seeking insects and the initiation of the disease on floral discs. Compare with Figures 7 and 9, representing artificial infections simulating inoculum dissemination by rainfall.

entirely confined to the blossoms, no twig blight having been found until 15 days after blight was detected in the blossoms, as may be observed in Table 4. Furthermore, the absence of frost injury made it possible to trace infection courts on the blossoms much more readily than is possible when blight symptoms are complicated by low-temperature injury. In numerous flower clusters the first blight symptoms were found to involve only the upper portions of the blossoms, Figure 12, clearly suggesting that the infections had started either on the receptacle or on structures borne on the receptacle. In all such cases, the leaves subtending the floral clusters as well as the lower parts of the pedicels were entirely free from disease. Unquestionably the earliest agent involved in disseminating the pathogen in this season was one that operated only in the floral tissues. Obviously rain or wind cannot be charged with such refined discrimination, and the evidence previously presented showing that water-borne inoculum applied to flower clusters produces infections indiscriminately on leaves as well as floral parts, and on upper parts of flowers as well as on lower portions of pedicels, is seemingly sufficient to exclude meteoric water as the disseminating agent for the first infections of 1929. It may be noted that

these infections preceded infections on Transcendent and Whitney crab-apple varieties by about one week. These varieties stood in the same orchard as the Kieffer and Winter Nelis pear varieties, which showed the earliest infections.



Figure 13. Compound infection involving both flowers and leaves of Kieffer pear, typical of natural infections in which the disease has been operative a much longer period than in infections not retarded by frost-injury. Photographed May 4, 1931, following a March freeze which killed fully 95 per cent of the pear blossoms.

It seems desirable to emphasize the hazard of attempting to determine infection courts for primary infections after the disease has been operating for some time. When the malady involves both leaves and blossoms attached to the same spur (see Figure 13), as it frequently does, then it is clear that attempts

to trace the origin of the infection are made under exceptional difficulties. Even where leaf infections are not to be found independent of flower infections, there is no certainty that the disease may not have originated in leaves attached to flower clusters rather than on upper floral parts. On the other hand, such compound infections do not necessarily lend support to the idea that infections started on the leaves. An inspection of Figure 13 reveals that in this instance the disease involved the whole of each blossom, the leaf petioles, and the laminar tissue surrounding the midribs, particularly in the basal portion of the leaves. The tissues which showed the most discoloration and the greatest amount of collapse and withering were the blossoms, including the pedicels, as may be seen in the irregular contour of the pedicel of the flower to the right. This, however, cannot be considered as conclusive evidence for the initiation of the disease on floral tissues, though it may be suggestive of such an act.

Not all flower infections are followed by leaf infections, in which the pathogen travels downward within the tissues of pedicels into spurs and leaf petioles. It is a peculiar and unexplainable fact that certain apple varieties, such as Grimes Golden, and even Kieffer pears at times show marked resistance to the passage of bacteria from blossom to spur and leaf tissues, (Figure 14). Now as far as the Grimes Golden apple is concerned, it has frequently been reported as very susceptible to both blossom and twig blight in various states. In Arkansas it rarely shows such susceptibility, although it occasionally is found to be seriously blighted. In perhaps most instances, however, a few blossoms will be found blighted without any adjoining leaf blight or independent twig blight. Why a variety, which is very susceptible in sections of the country where blight as a whole is not nearly as common or as destructive as in Arkansas, should be more or less resistant here, constitutes one of the most interesting and outstanding questions concerning resistance and susceptibility. It is of course not intended to suggest that all floral infections, in the absence of leaf or twig infections, are due to a difference in susceptibility of the different organs. In most instances the preponderance of evidence indicates that instead of differences in susceptibility between these parts, the reason for a differential infection in favor of blossoms is to be found in the availability of certain floral parts for infection, and the action of contaminated insects which restrict their activity to blossoms.

Briefly summarizing the 6 year data concerning first infections on pears in Arkansas, it appears that when frosts or low temperatures injure the blossoms, the first infections are to be found on both flowers and leafy shoots, with the former predominating. When no such injuries occur, primary blight is restricted to blossoms, and when it is detected early, it is not accompanied by leaf infections.



Figure 14. Flower and fruit infection of Kieffer pear, photographed May 4, 1931, in which the tissues of spurs seemingly show signs of resistance to the passage of the pathogen from flower or fruit. Note absence of infections on leaves, and the infections on fruit which measured approximately one-half inch in diameter, indicating that, although the disease involving the upper flower cluster was not observed until the malady was detected on the lower fruit cluster, the disease was initiated on the blossom cluster sometime before it started on the fruit. This may be interpreted as indicating primary infections on blossoms and secondary infections on fruit.

FIRST BLIGHT ON APPLES

Despite the fact that apples in Arkansas start growing later than pears, thus frequently escaping spring frosts which are destructive to the latter, it is even more difficult to trace the origin of the first blight on apples than on pears. There are two principal reasons for this. First, growth, including blossom production, coming later than on pears, makes it possible for the disease to be initiated on the latter and then be spread to apples; when this occurs the blight on apples is of course not primary blight⁷ but secondary. Second, the very hairy growth on all common susceptible varieties of apples grown in this state interferes greatly in detecting early blight symptoms, and in large measure prevents a clear distinction between infections which may have been initiated a number of days apart. For these reasons the writer does not feel warranted in unqualifiedly accepting his own observations on assumedly primary blight in apple orchards which were not outside of at least a 3-mile radius of pear trees, or of accepting the findings of other investigators who have not attempted to determine the presence of blight on early blooming pears, on crab-apples where these are commonly grown, on cultivated hawthornes, or on other early blooming susceptible plants. A study of fire-blight literature, particularly the more recent publications, reveals few instances in which the investigators have attempted a thorough survey of all susceptible plants within a 3-mile radius of the orchards which they investigated, with the result that in many instances there is no assurance that the blight which is taken to be primary is not secondary. The practically unanimous belief among plant pathologists that all blight is initiated by the activity of local hold-over blight has tended to limit studies to a few orchards in a restricted area.

With these limitations in mind, it seems doubtful to the writer whether any large proportion of the earliest apple infections in orchards of Washington and Benton counties can with certainty be listed as primary blight. Even in years when the whole pear crop is entirely destroyed by frost or winter freezes, a common occurrence, there is no absolute assurance that some of the pear blossoms had not been inoculated with fire-blight bacteria prior to their severance from the spurs, and had not been visited by insects which later distributed the bacteria to apple blossoms. Again, since no infectious exudate originating in hold-over blight has been found in the early part of the growing season, the determination of the source of inoculum, and the proper designation of the first blight, whether primary or secondary, becomes a maze of uncertainty.

⁷By primary blight is meant here the first blight engendered by overwintered material.

Out of the several hundred apple orchards which have been under observation, there is only one that the writer considers sufficiently isolated to warrant its classification as a producer of primary blight. This orchard is not located in the main fruit belt, but in Franklin County, about 10 miles north of Ozark, and about 60 miles from the main apple growing section. It is situated near the summit of a mountain, in a very rugged, hilly section that is sparsely populated and that is mostly in natural forests. The only cultivated fruit trees in the vicinity of this orchard are located about 3 miles to the east. The latter trees form a fair-sized apple orchard which was found to be free from blight when the orchard to the west was severely blighted. No blight having been found within 10 miles or more of the west orchard, it offered an exceptional opportunity of observing the sequence of blight, without the complication of outside blight playing a part. The orchard contains two blocks of Jonathans separated by about four rows of non-susceptible apple varieties. Each block of Jonathans consists of around 200 22-year old trees, the soil of one being decidedly superior to that of the other, resulting in a considerable difference in growth of the trees. Contrary to its usual behavior, blight over a series of years has been almost entirely restricted to the block in which the trees had made the poorest growth. This is so different from most cases which have come under the writer's observation, and so different from the commonly accepted ideas concerning the relationship of blight to vigor and rate of growth, that it constitutes an enigmatic exception. It recalls a similar observation made by Whetzel and Stewart (50, p. 34) about 24 years ago, in which they found poorly growing pear trees "standing in sodded soil of a stony hillside" being killed by blight in a single season.

During the three consecutive years in which this orchard has been under observation, the first signs of blight were always confined to the blossoms, and the twig blight which developed later could invariably be traced to the previous blossom infections. In 1930 and 1931, the two recent years in which blossom blight was very extensive in this orchard, the blossom infections appeared simultaneously over the whole susceptible block, and in numerous instances one or more blossoms in a cluster were at first found blighted when the remainder of the blossoms in the cluster and when the leaves arising from the same fruiting spur showed no signs of blight. So severe has blossom blight been in the susceptible block that in some years, as in 1930, it resulted in almost complete killing of the apple blossoms, most of the trees having nearly all their blossom clusters killed. Every limb which possessed blossom clusters in 1930 was blighted, in sharp contrast to the non-blossoming branches which remained free from blight until 7 to 10 days after blossom blight had appeared. Indeed in the years 1930, 1931, and 1932, most

of the twig blight involved succulent shoots which arose from blossom-bearing spurs, and the shoots that appeared from terminal or lateral leaf buds were as a rule free from blight. In other words, most of the twig blight seemingly consisted of internal extensions from blighted blossoms, while twig blight, unaccompanied by blossom blight, was negligible as a whole. In addition, the downward invasion by the pathogen of older wood from a single blighted blossom cluster rarely involved more than a few inches and rarely reached wood that was more than a half-inch in diameter. This gave the older and larger branches an aspect of complete resistance, totally unlike that of the younger, newer growth involving the flowering wood.

The extreme amount of killing of the bearing wood over a series of years, has resulted in a much stunted appearance of the trees, with thick scaffold branches bearing short secondary limbs, and forming small but open tops. This has further resulted in direct exposure of the larger limbs and trunks to the sun, culminating in large dead areas of the bark on the upper side of the limbs and on the south side of the trunks. When first observed, these dead areas were taken to be fire-blight cankers, but as no infections were obtained from the living and dead wood taken from the borders of the affected areas, as they mostly existed on the sides directly exposed to the sun, and as no bacterial ooze was observed issuing from them, they are more likely to fall into the type of injury known as sun scald. Often these areas were found to harbor various wood-destroying fungi, such as *Schizophyllum*, which were evidently increasing the sizes of the dead bark areas and attacking the underlying wood.

If the pathogen overwintered at all in this orchard, it must have done so in conjunction with the blight originating in flower clusters and small twigs. The almost complete absence of blight on limbs larger than one-half inch in diameter, necessarily means in this instance that hold-over cankers, in the sense of large limb or body cankers, could not be held responsible for the overwintering of the fire-blight bacteria. Furthermore, the widespread and apparently simultaneous distribution of blight over all blossoming trees in the susceptible block in the epidemic years, makes it difficult to assume that overwintered blighted twigs and blossom clusters were responsible for the initiation of blossom blight. Since late winter pruning has been a regular practice in this orchard, the former year's blight which escaped pruning was necessarily limited in amount, and more or less scattered. It is more in keeping with the observed facts to assume either that the blight bacteria lived through the winter superficially on both diseased and healthy wood and buds, being spattered and otherwise contaminated with bacterial masses involved in the exudates of the former season, or that the bacteria

were brought in by blossom-visiting insects which harbored the pathogen.

The last explanation seems more reasonable at present; otherwise it would also be necessary to postulate that the blossoms were the only susceptible organs on the trees, and that the young leaves were entirely resistant.

As in the main apple-growing sections of Arkansas, no infectious exudate was found in this isolated orchard in the early part of the growing season prior to the first appearance of blight. Each year a careful search was made on all blighted trees in the early spring for signs of oozing from formerly blighted tissues. A few cases were found in which droplets of exudate were located in or near the margin of last year's blight, but in every instance this exudate when diluted with sterile water and used for a series of dilution plates and for direct inoculation into tender pear twigs failed to produce infections. This evidence taken in conjunction with the fact that in at least 1930, when no rainfall occurred throughout the entire blooming period and when blossom blight was extremely common in this orchard as in many other Arkansas orchards, suggests that the bacteria were brought in by blossom-visiting insects. Further than this, the extreme isolation of this orchard makes it reasonable to assume that the blight here was of the nature of primary blight unconnected with earlier blight on trees existing outside of this orchard. The only wild plants found growing in this region that may be susceptible to blight are species of *Crataegus*, and although a careful search was made for the disease on these wild plants none was found.

Concerning blossom-visiting insects that may have been involved in initiating blossom blight, it is to be noted that in addition to wild bees, a relatively large, though somewhat neglected apiary is located within one mile of this orchard. There is no shortage of bees here at blooming time.

Whether or not the first blight on apples in the main apple-growing section of Arkansas is primary or secondary, it is almost always in the nature of blossom blight. Table 4 shows that during the last 6 years, the first blight was blossom blight exclusively in four of these years, and in the other two, 1931 and 1932, the years of late spring freezes, it consisted of both blossom and twig blight. In 1931, the March freeze had injured apple blossoms seriously in numerous orchards, and to the extent that the disease was not detected until 2 to 3 weeks later than usual, while in 1932 a single blighted shoot was found at the same time as many blossom clusters were detected with disease, and in this single case the two inches of intervening tissues between the shoot and a blighted blossom cluster were found to be surcharged with bacteria, the latter producing the

characteristic milky clouding of fire-blight bacteria in water mounts. It has been recorded already in connection with the discussion of first blight on pears that in 1931 the fire-blight pathogen was isolated from frosted apple blossoms which were about to fall, and which showed no outward symptoms of blight except droplets of bacterial exudate. It is, therefore, probable that in these 2 years the first blight was blossom blight as in the 4 years previous.

Not only were the first blight symptoms to be found restricted usually to the blossoms, but on these parts the disease was not detected until after petal fall. The earlier report by the writer (27, p. 68-70) of fire-blight infections being found on closed blossoms was due to the failure to distinguish between two different bacterial diseases present on pomes in Arkansas. More recent work by the writer jointly with W. L. Bleecker (32) has shown that in addition to fire blight, another bacterial disease is present particularly in the early part of the growing season, which presents symptoms on leaves, flowers, and fruits that are difficult to distinguish from fire-blight symptoms. Furthermore, the pathogen presents such numerous morphological, cultural, and physiological reactions similar to those shown by *Er. amylovora* that it was necessary to employ serological tests in order to be certain that it was not the same species. While it is entirely possible that first blight may be found in some sections of the country on closed blossoms as well as on leafy shoots, as investigators in Wisconsin have reported (2, 20), it is also possible that this other disease, termed blast, has been mistaken at times for fire blight.

PRIMARY TWIG BLIGHT ON APPLES AND PEARS

Although much of the twig blight on pears and apples is secondary blight which is directly or indirectly connected with blossom blight, there is another type of twig blight which shows no such connection. It has in all instances been found on trees or close to trees which were blighted in the previous year and in some cases it has involved trees which currently had borne no blossoms. There is evidence to show that it may involve four different methods of overwintering of the pathogen and three kinds of infections: first, infections by means of internal extensions of previous year's blight; second, bud infections of the previous year; third, infections induced by bacterial exudate of the previous year which remained alive through the winter on the exterior of twigs and limbs; and fourth, infections brought about by bacterial exudate from hold-over cankers on relatively large limbs and trunks.

Definite evidence for the first of these methods has been found on water sprouts that became infected in the past season.

and in which the bacteria continued to live within the tissues and to invade for considerable distance either the uninfected portions below the terminal blight of the past year or new shoots that had been formed on the old wood near the previous year's blight. This blight has mostly been found after blossom blight had been in evidence. In no instance has it been observed in abundance, not more than one or two shoots for every 25 trees. In each case the external appearance of the sprout indicated that infection had occurred during the middle or latter half of the growing period of the previous season, after the sprout had made a growth of two to three feet; the infections, presumably occurring near the tips, had run downward for a foot or more, then as growth slackened, they remained stationary for the remainder of that season. The following spring, as conditions became proper for growth of both sprout and bacteria, the latter continued their downward invasion. A microscopic study of the internal tissues of the old and new infections seemingly confirmed this view; the bacteria were found in continuous strands reaching from the previous year's blight to that of the current year, as was observed in histological sections made on a sliding microtome. However, this was not confirmed by pure-culture isolations in these instances. The writer (29, *Figs. 11, 12, 13*) has previously called attention to the possibility of such internal extensions of the previous season's blight resulting in new twig blight, and Miller (20) has apparently observed the same thing in Wisconsin apple orchards.

While numerous writers have previously called attention to the renewal of activity of overwintered cankers and blighted twigs, they have in the case of twig blight associated this mainly with the production of spring exudate which served as inoculum for primary blight, as both Brooks (2) and Pierstorff (23) have reported. Brooks noted some blight extension beyond the margin of the previous year's blight, but this he evidently found to be more or less localized, as nothing is said about any considerable amount of fresh internal invasion either into older wood or into new shoots formed on the old wood near last year's blight.

The fact that internal extension of bacterial strands may produce new twig blight in amount comparable to that of the previous season, either on the same twig or on a new shoot arising from it, apparently has not been described previously, although there is some described evidence to the effect that overwintered blight on large limbs may result in continued and more or less extensive invasion in the next season. Fundamentally the difference between those twig infections previously reported and those here described is that in the former the new infections occurred externally by means of the bacterial exudate, while in the latter they occurred internally by means of renewed invas-

ions from old infections. In the last instance the exudate appears after the new infections have developed sufficiently that the amount of new bacterial growth, produced simultaneously with the new growth of the host tissues, is large enough to permit the forcing of the bacteria out of the tissues. In the first type, the exudate presumably produces primary blight and in the last type it produces secondary blight, if by primary blight is meant the first new blight that is produced from overwintered material. This distinction, however, is capable of being exaggerated. For example, a body canker or a large overwintered limb canker may extend its dimensions in the following spring and at the same time produce exudate which cannot be attributed wholly to last year's blight.

While such pear twig blight has in almost all instances been observed after blossom blight had appeared, and in one case simultaneously with earliest blossom blight, there still is the possibility that it may, at least in some portions of the United States, appear early enough to act as sources of inoculum for blossom blight. Also, the same thing may occur on apples, although this has not been clearly verified up to the present. The most important consideration with reference to this form of blight is that it is dependent neither on insects nor on rain for its production, save perhaps indirectly in rendering the tissues susceptible to internal advance of the pathogen by the absorption of sufficient moisture.

The second method mentioned in primary twig infections, the one involving bud infections of the previous year, has not yielded conclusive evidence up to the present. The investigations which point to this possible method of overwintering and infection consist of the following: It has not infrequently been noted that buds located within or near blighted areas on pear twigs and limbs, in which the blight had not entirely killed the branch and in which the buds remained alive, bore at times discolored areas at the base of the outer scales. Were such discolorations due to the presence of *Er. amylovora*, and if so could they serve as a source of overwintering and blight production in the following year?

To answer these questions several hundred Kieffer pear buds were gathered in the late fall and winter, carefully severed from the branches so that no limb tissues adhered to the bud, surface sterilized with a solution of mercuric chloride, mashed in Petri dishes containing sterile water, and injected by means of hypodermic syringes into healthy pear shoots maintained in a greenhouse. One of these buds gathered at Bentonville, Arkansas, October 8, 1921, produced typical fire blight infections. From one of the pear shoots which became diseased *Er. amylovora* was isolated, grown in pure culture from single-colony transfers

and when again inoculated into healthy pear shoots reproduced typical blight symptoms. The original bud was located within a blighted portion of a vigorous shoot, and when gathered it was alive and appeared perfectly sound save for a brown discoloration at the bases of the outer scales. A microscopic examination revealed blight bacteria present not only in these scale bases, but also in portions of the interior of the bud. Judging from the internal and external appearance of this bud, there is good reason to believe that it probably would have grown the next spring. Assuming that the bacteria within the bud would have remained alive, not a very hazardous assumption, blight probably would have appeared on the shoot originated by this bud. The shape and size of the bud indicated that it was vegetative and not flower-bearing. It is obvious that this finding requires verification and amplification. It is, however, sufficient to indicate another possible source of overwintering and infection, although Stevens and his associates (37) have presented negative evidence in the form of artificial bud inoculations by applying water suspensions of *Er. amylovora* to pear buds.

The third method postulated for the production of primary twig blight, from bacterial exudate produced in the previous growing season, is being investigated very thoroughly, and the report here presented is largely in the nature of a preliminary announcement. While there are a number of investigators who have reported that *Er. amylovora* when exposed to the sun and air perishes in relatively short time, there are two or three who have found that this pathogen in the form of exudate may remain alive for a number of months when kept under indoor laboratory conditions. For the present the writer wishes to record briefly that he has succeeded in obtaining typical fire-blight infections and pure cultures of *Er. amylovora* from blighted apple and pear twigs gathered in midwinter and soaked for a short period in sterile water. In one case the cut ends of the twig were coated with paraffin to avoid the possibility of bacteria coming from within the tissue contaminating the water in which the twig was soaked. This soaking may be compared with a layer of rain-water wetting a surface on which bacteria are present.

These three methods of overwintering and manner of producing primary twig blight are all additions to the one well-recognized method. The latter, involving exudate production and extension of the previous year's blight on relatively large limbs or trunks, is also operative in Arkansas, but the production of exudate seemingly is at an entirely different time in the growing season than has been found in other sections of the country. Here it does not usually make its appearance until late in May or in June, which in period of growth, would correspond to the end of June and July in more northern states. Consequently, it is not in evidence until after blossom blight has

been well established, after one or more of the other types of primary twig blight has appeared, and frequently after secondary blight has appeared. On apple varieties, such as Jonathan, Yellow Transparent, and Maiden Blush, this method of overwintering usually occurs in connection with limbs that are one to two inches in diameter, and is mostly confined to those orchards, which because of richness of soil, fertilizer applications, or other factors that are associated with increased susceptibility, develop blight each year. This method of overwintering and production of primary twig blight is often found in association with cone-shaped infected portions of trees, or concentrated amounts of blight below and around the hold-over canker.

The relatively large limb or body cankers which are concerned in this form of overwintering, produce at times copious flows of bacterial exudate. When this occurs, the exudate instead of taking the form of rounded, well defined, and very adhesive drops, partakes more of the nature of a watery sap flow. It is, therefore, much more readily washed down and blown about by rains and winds than the viscid, tenacious drops commonly formed on smaller blighted twigs and flower clusters. Indeed, the flow at times is of such reduced viscosity and so abundant that without the aid of rain or wind, it reaches twigs and limbs which are one or more feet below the canker, and which happen to be in the path of the flow.

Briefly summarized, it appears that primary twig blight in Arkansas is not ordinarily in evidence until blossom blight has had ample time to develop, and that it may arise at different times in one of four different ways. With this in mind, it is clear that overwintering is much more of a complex problem than has previously been postulated in the literature, and that control measures which do not take all of these factors into consideration are likely in some cases to prove inadequate. It is also clear that depending on the source of the primary inoculum and the time of its dissemination, control measures which may be successful in a given region or in some orchards within that region, may prove to be failures in others.

INSECTS AS CARRIERS AND DISSEMINATORS OF THE FIRE-BLIGHT PATHOGEN

INVESTIGATIONS ON HONEYBEES

Having found that the first blight in Arkansas in the greatest number of cases involved blossoms of both pears and apples, and having failed to correlate such blossom blight with the activities of overwintered cankers and blighted twigs, and at times with periods of rainfall, the questions which naturally presented themselves were, what is the source of inoculum for this first

blight, and what agents are involved in spreading this inoculum to the blossoms?

The excellent work on the relationship of rainfall to fire-blight dissemination in Wisconsin (20) and Michigan (41) seems to leave little doubt that in those states overwintered cankers and blighted twigs serve as the source of inoculum for first blight, and that this blight is correlated with periods of rainfall. While there is fairly good evidence to show that secondary blight in this state is in large measure due to bacterial dissemination by rain and wind, as far as the first blight is concerned, the findings in Wisconsin and Michigan bear no evident resemblance to those in Arkansas. Aside from the question as to the source of inoculum, it has already been shown quite fully that in some seasons, as in 1930, when there was no rain during the entire blossoming period of apples, blossom blight was much more common and far more destructive than in years of ample rainfall. As far as activity of overwintered blight is concerned and the serving of such blight as the source of inoculum for the first blight, it has also been recorded that in this same year as well as in others, the disease was abundant in orchards and groups of orchards which possessed little or no blight in the previous year, clearly indicating that overwintered blight played no part in these orchards, and that the pathogen must have been brought in from some outside source. Furthermore, this outside source, if overwintered blight was its origin, in many instances was not found to be closer than one to several miles. Irrespective of the source of inoculum, it is evident that some agent or group of agents which possesses the ability of covering considerable distances and which on the growing plant confines its activities to blossoms, was involved in disseminating this first blight. As far as the honeybee is concerned, von Frisch (5) reports that on level or slightly hilly country, it will travel as much as 3 to 4 kilometers, 1.86 to 2.48 miles.

In a preliminary article (30) the writer has discussed the remarkable pioneer work of Waite, who showed that honeybees became contaminated with fire-blight bacteria when they visited blossoms which he artificially inoculated. He also found that such bees were capable of spreading the disease to other blossoms. From this he at first assumed that bees as well as wasps and flies obtained the inoculum under natural conditions from bacterial exudate originating in hold-over cankers. In most of his later publications, however, he apparently gave up the idea that bees are distributors of primary inoculum. For example, in one of his later publications (45), after describing the production of bacterial exudate on hold-over cankers, he reports that this gummy exudate is "visited by insects, especially wasps and flies, and these carry the germs on to the newly opened



Figure 15. Artificial infection on Bartlett pear shoot induced by the injection of a water-dilution of about 15 grams of honey + honeycomb gathered from a beehive located in a blight-infested apple orchard. Injection made on Aug. 6, 1929; photographed Aug. 24, 1929. *Er. amylovora* was isolated from this infection, reinoculated into pear shoots, and typical infections produced.

blossoms, and bees and other flower-visiting insects continue the work of distributing it." In other words, Waite considered the honeybee as a distributor of secondary inoculum. This is in harmony with the conclusions of Gossard and Walton (6). These investigators claim to have failed to find the fire-blight pathogen within beehives in the early spring. Two different methods were used to determine this point, the plating-out method with honey and wax, using "dozens of samples", and direct inoculations with drops of honey into wounded apple shoots, the latter consisting of 670 inoculations. The first method failed to yield any cultures of the fire-blight pathogen. The second method, with samples of honey from three hives, resulted in the death of 28 per cent of the twigs in one series of inoculations, 32 per cent in another series, both from one hive; 12 per cent in one series from another hive; and 26 per cent in one series and 48 per cent in another series from a third hive. The last hive was located under a cloth-enclosed apple tree upon which the pathogen had been introduced artificially. The first two were open hives existing under natural conditions. Since only one check twig became blighted, out of several hundred, it seems to the writer that contrary to the authors' conclusions, the evidence is quite substantial that the fire-blight pathogen was obtained in a relatively large number of cases from samples of honey taken from the two hives which were beyond the reach of the artificial inoculum. Most unfortunately they fail to give the details of the 473 inoculations attempted with honey from these two hives, particularly the dates of application and the number of series used, although it may be inferred from a general statement made concerning this work and the work pertaining to inoculations from pollen-baskets and mouth parts of bees, that May 23 was the date for all successful inoculations with samples of honey. As "samples from all three hives were taken and used daily throughout the entire blooming period," nothing being said about sampling prior to the blooming period of apples, there is, of course, no assurance that the infections obtained were not derived from honey which became contaminated from bees which visited earlier infected blossoms of the current season. This objection, however, is not proved by any data which they present, and their statement (*p. 90*) that "so little blossom blight was present anywhere in the orchard that we concluded no results would be forthcoming" seemingly suggests that it may not be valid in this instance. The important point to the writer is that, despite Gossard and Walton's statement that they "were unable to obtain cultures of fire blight from hives in early spring" their data involving direct inoculations with honey lends little or no support to such conclusion. The fact that they were unable to isolate the pathogen by the plating-out method is not at all surprising, as will be shortly demonstrated.



Figure 16. Artificial infection by means of a pure culture of *Er. amylovora* isolated from a blighted Bartlett pear shoot, the disease being induced by the injection of a water-dilution of about 20 grams of honey + honeycomb + pollen cells. Material gathered from a beehive located in a blight-infested apple orchard, on Nov. 9, 1929. Infection made on Nov. 25, 1929; organism isolated a few days later, reinoculated into healthy pear shoots, and infections again produced. Photographed Dec. 13, 1929.

Concerning the number of blossoms capable of becoming infected by the visitation of a contaminated bee, Miller (20, p. 595) has presented some interesting information. By the use of a honeybee proboscis attached to a stick, he found, after dipping the proboscis into the nectarial surface of a diseased pear blossom and then dipping it into the nectar of 15 healthy pear flowers, that every one of the flowers thus inoculated became diseased in a characteristic manner. He suggests that pollinating insects may still be potential agents of dissemination even after having visited 15 flowers following contamination.

Although Gossard and Walton concluded as a result of transferring pure cultures of *Er. amylovora* to sterilized honey, that the organism can live only a few days in honey, not much more than 72 hours, McClarty (16) found that in some of his transfers of the fire-blight pathogen into strained honey contained in test tubes, the bacteria remained viable from January 9 to February 26, a period of 48 days. A few months after the writer's preliminary announcement concerning the isolation of the germs from beehives, Thomas (40) reported that when a heavy suspension of *Er. amylovora* in water was applied to the surface of a frame of honey, to the surface of the comb, and to uncovered cells at the margin of the comb, the organism was recovered from the wooden frame up to 20 days after transfer, from the surface of the comb up to 55 days, when the experiment was terminated, and up to 15 days from the uncovered cells. A second trial, in which undiluted pear-fruit exudate was applied to the surface of the comb only, cultures were obtained up to 35 days after transfer, when the experiment terminated. He also reported that he succeeded in obtaining virulent organisms from bacterial exudate on apple twigs kept in the laboratory for 12 months at Ithaca, New York. The latter finding is quite in line with that of Waters (49) in New Zealand, who in 1922 recorded that fire-blight bacteria are capable of living in dried exudate for a period of 9 months. It is also comparable with the recent report by Pierstoff (23, p. 29) in which dried exudate on yearling Transcendent crab-apple trees kept in the laboratory, yielded cultures for the remarkable period of 2 years and 3 months.

In attempting to obtain information on the life of this organism in beehives, the writer began the work with the following in mind: First, the use of artificial transfers of bacteria into beehives, while offering interesting possibilities, cannot be considered of much value because there is no assurance that the type and quantity of inoculum, manner and time of distribution within the hive, and the medium upon which cultures had been previously maintained, will in any way compare with the nectarial mixtures, the manner of distribution on waxy surfaces or within the wax, the pollen mixtures, the enzymatic activities and types of dehydration which occur during the manufacture and

storage of honey, the mode of bacterial distribution by worker bees as they traverse brood cells, honey cells, and pollen cells, the time and manner of capping the different cell types and the relationship of this process to kind and amount of oxidation and evaporation, and numerous other activities which human hands cannot at present duplicate and which may have some influence on the life of any microorganism which is brought into the hive. Second, if any large number of overwintered fire-blight bacteria were present in any number of hives, or if any large number of bees present in the hives were contaminated in the early spring prior to the appearance of new blight, there is no good reason to doubt that, given suitable conditions for disease expression, whole crops over very wide areas would be destroyed annually. Again, considering the number and diversity of flowering plants visited by bees, it is obvious that any bacteria brought into the hive would eventually be very greatly diluted or obscured by the addition of nectars, pollen masses, and wax secretions. Obviously to attempt to find live fire-blight bacteria in a well stocked hive requires, even with the best technic at hand, a most arduous search. Third, the objections that have been offered from time to time relative to the improbability of fire-blight bacteria living within beehives because of the concentration of sugar in honey, because of its levulose content, and because of bees not visiting blighted blossoms or bacterial exudates, did not seem to the writer to be very weighty. It is quite well known that despite the concentration of sugar and levulose content, numerous live microorganisms are present in honey. In addition to honey cells, there are so many other cells and places in the hive that may become contaminated that even if honey possesses properties that are germicidal to *Er. amylovora*, this is not sufficient to exclude the whole hive from consideration, not even that part of the comb which contains honey. As to bees not visiting blighted blossoms or bacterial exudates, while it may be true that bees would not visit blossoms after they are blighted, this does not warrant the assumption that they would not visit blossoms which contain bacteria in the nectar but which had not as yet developed blight symptoms. Waite, for instance, has in numerous publications reported that these bacteria are capable of multiplying in the nectar, and this has been confirmed by Gossard and Walton (6), who in addition showed that *Er. amylovora* may live and be infectious for at least 5 days in the nectar of blossoms which are not known to be susceptible to fire blight, as for example, peach blossoms.

Having located an orchard in the apple section which showed considerable blossom blight and in which a number of beehives were maintained, the first effort was to develop a technic and to standardize the methods sufficiently so that any microorganism isolated could be with reasonable assurance ascribed to



Figure 17. Artificial infection at base of Bartlett leaf-cluster, obtained by injecting a water-dilution of honey + comb gathered Dec. 17, 1929, from a beehive located in a blight-infected apple orchard. Injection made Jan. 8, 1930; first signs of infection appeared Jan. 20, 1930. Organism isolated from this infection, reinoculated into pear twigs, and infections again produced

the samples taken from the hives, and also to enable the detection of any fire-blight bacteria that may be present. With reference to the last point, it may be noted, however, that the writer is in doubt, after 3 years or more of this work, concerning the invariable detection of *Er. amylovora* in beehive samples by the methods utilized.

The technic which was finally developed and used throughout the work was as follows: Having found that half-pint cardboard containers such as used in the ice-cream trade, could be easily sterilized in an autoclave at 15 pounds pressure for 30 minutes without injuring them, and that they could be handled with greater ease and less breakage than Petri dishes or test tubes, these were utilized for gathering the beehive material. An alcohol lamp was carried out to the field, together with the equipment necessary for working with beehives, and the knife used for gathering the material was flamed before and after each sample was taken. When bees were gathered, they were similarly handled with aseptic precautions. Having filled a container with samples taken from various parts of a hive, the lid was immediately put in place and the number of the hive and the date of sampling written on the lid. If any sample was not tested immediately after it was gathered, it was placed in an electric refrigerator, maintained at a temperature of around 10°C. When live bees were gathered, the container was always placed in the refrigerator to quiet them and permit ready handling of relatively large numbers.

The writer's first attempts to isolate the fire-blight pathogen by means of the plating-out method and by placing beehive samples into tubes of nutrient broth were all failures. Invariably it was found that such common saprophytes as species of *Aspergillus* and *Penicillium* and rapidly growing, non-pathogenic, bacterial colonies were present in such great numbers on all samples, that there was practically no possibility of isolating the relatively small and somewhat slow-growing colonies of *Er. amylovora*. The method finally chosen was as follows: All the preliminary work with the samples was done in a closed, heavily steamed, transfer chamber, and all instruments used for handling the samples were sterilized in an open flame prior to their use. The hands were carefully washed, rinsed in a solution of mercuric chloride, and allowed to dry without removing the disinfectant. About 10 cc. of sterile distilled water was poured into each sterile Petri dish and a portion of honeycomb, honey, pollen cells, brood cells, or live bees from each field sample was mashed and thoroughly distributed in the water. The amount of beehive material used in each dish varied somewhat and no effort was made to obtain equal weights, a procedure which would have meant exposing the samples to laboratory air and dust. However, the weight of the material placed in each dish was compar-

able and approximated 10 to 15 grams. No effort was made to separate the honey from the comb, the pollen from the pollen cells, the larvae from the brood cells, or in most instances, the mouth parts of live bees from the remainder of the bodies, although in a few instances this was done. In each case the material as it came from the hive was treated more or less as a unit, an empirical but seemingly essential procedure in such a large and complicated task.

At least three injections into healthy, succulent Bartlett pear shoots were made from each dish, using sterilized hypodermic needles attached to sterilized syringes. All the injections were made on potted pear trees growing in a greenhouse. In all instances a sufficient number of controls were maintained in the form of uninoculated shoots or of shoots injected with sterile water. The number of contaminations which occurred on these checks was surprisingly small, considering the crowded conditions of the greenhouse, and in the whole 3-year period in which these tests were conducted, involving many hundreds of injections, the total number of contaminations comprised four blighted leaf clusters, and in each case the concomittant injections were disregarded and do not appear in the final records. Before this work was initiated the writer had observed in the previous 3 years experiments with fire blight in the greenhouse that when overhead watering was avoided there were no contaminations, but when the tops of the trees were subjected to daily sprinkling, the disease was likely to spread to uninoculated shoots if any blight was present in their vicinity. The few accidental contaminations obtained were in each instance apparently traceable to lack of vigilance in applying water to the plants. Not only were all infections rejected when any accidental contamination was present, but unless infections centered immediately around the point of injection, they were not considered as being due to the applied inoculum. Furthermore, unless the infection appeared in less than 3 weeks after the inoculation, it was likewise rejected. While this practice appears somewhat rigid in the light of other data indicating that external signs of infections may at times be held in abeyance for over 3 weeks after inoculation, depending on growing conditions of the host, on amount of inoculum, and on other factors, nevertheless, it appeared justified in order to be certain that if errors were made they would be on the safe side.

When an infection showed typical blight symptoms around the point of injection within a few days after inoculation, and when no accidental blight occurred on controls, the infected material in each instance was surface sterilized, macerated in sterile water, and used for a series of poured dilution plates in the usual manner. If bacterial colonies appeared, transfers were made from single colonies and used for another series of injec-



Figure 18. Artificial infection on Bartlett shoot induced by a pure culture of *Er. amylovora* isolated from a blighted Bartlett pear shoot, the disease being induced by injecting a suspension of the mouth-parts of 10 honeybees macerated in sterile water. Bees gathered on Feb. 8, 1930, from a beehive located in a blight-infected apple orchard; injection made Feb. 10, 1930. The first signs of infection appeared Feb. 28, 1930. Organism plated out a few days later and single colony transfers produced characteristic symptoms on pear shoots. Photographed March 7, 1930.

tions. If these in turn resulted in blight infections and no contaminations occurred in the controls, the evidence was considered adequate. The number of trials, and successful infections with beehive material are recorded in Table 5.

TABLE 5

Experi- ment No.	Material gathered	Date of injection	No. of injections	Nature of material	No. of infections	Date of infections
	1929	1929				
1	June 27	June 28	40	honey + comb -----	0	-----
2	July 6	July 6	30	honey, comb, pollen --	0	-----
3	July 15	July 15	60	honey, comb, brood cells & young larvae	0	-----
4	July 29	Aug. 6	66	honey + comb -----	3	Aug. 24
5	Sept. 21	Oct. 9	54	honey + comb -----	0	-----
6	Nov. 2	Nov. 25	75	honey + comb + pol- len cells -----	2	Nov. 28
7	Nov. 9	Nov. 30	60	honey + comb -----	0	-----
8	Nov. 9	Dec. 5	30	honey + comb -----	0	-----
9	Dec. 17	Dec. 19	60	live bees + honey, comb, pollen cells --	0	-----
10	Dec. 17	Dec. 21	60	honey + comb -----	1	Dec. 28
	1930					
11	Dec. 17	Jan. 8	40	honey + comb -----	2	Jan. 20
12	Dec. 17	Jan. 20	10	honey + comb -----	2	Feb. 1
	1930					
13	Feb. 1	Feb. 3	150	honey + comb, live bees -----	0	-----
14	Feb. 1	Feb. 6	100	honey + comb -----	0	-----
15	Feb. 8	Feb. 18	75	live bees, honey + comb, pollen cells --	0	-----
16	Feb. 8	Feb. 10	102	mouthparts of 10 bees (Total No. bees used 493)	1	Feb. 28
17	Feb. 8	Feb. 11	30	mouthparts of bees, honey + comb -----	0	-----
18	Feb. 8	Feb. 14	75	honey + comb, pol- len cells -----	0	-----
19	Feb. 27	Feb. 28	30	120 live bees -----	0	-----
20	Feb. 27	Mar. 1	120	841 live bees -----	0	-----
21	Mar. 13	Mar. 14	54	227 live bees -----	0	-----
22	Mar. 13	Mar. 15	42	348 live bees (mouth parts) -----	0	-----
23	Mar. 27	Mar. 28	108	794 live bees (whole) -	1	April 12
24	Apr. 8	Apr. 10	78	407 live bees (whole) -	0	-----
25	July 9	July 11	120	honey + comb -----	0	-----
26	July 15	July 15	75	honey + comb -----	0	-----
27	Aug. 6	Aug. 7	165	honey + comb -----	0	-----
28	Aug. 9	Aug. 11	150	honey + comb -----	0	-----
29	Sept. 28	Sept. 29	5	honey + comb -----	0	-----
30	Oct. 28	Oct. 28	30	honey + comb -----	0	-----
31	Nov. 25	Nov. 25	30	honey + comb -----	0	-----
32	Dec. 17	Dec. 17	33	honey + comb -----	8 ¹	Dec. 19 to Dec. 21
	1931	1931				
33	Jan. 17	Jan. 17	84	honey + comb -----	0	-----
34	June 12	June 12	15	honey + comb -----	1	June 20
35	June 29	June 29	20	honey + comb -----	0	-----
36	July 6	July 6	25	honey + comb -----	0	-----
37	July 6	July 7	5	honey + comb -----	0	-----
38	July 22	July 22	19	honey + comb -----	0	-----
39	Aug. 6	Aug. 6	15	honey + comb, pol- len cells -----	0	-----
	1932	1932				
40	Feb. 22	Feb. 23	24	honey + comb + pol- len cells -----	0	-----
41	Feb. 24	Feb. 24	18	honey + comb -----	0	-----

¹All infections obtained on Dec. 19-21 came from material gathered in one hive, the remaining 25 injections made at the same time with material from other hives yielded no infections.

TABLE 5 (continued)

Experiment No.	Material gathered	Date of injection	No. of injections	Nature of material	No. of infections	Date of infections
42	Feb. 25	Feb. 25	21	honey + comb + pollen cells	0	-----
43	Feb. 29	Feb. 29	15	honey + comb + pollen cells	0	-----
44	Mar. 1	Mar. 3	15	comb + slight amt. of honey	0	-----
45	Mar. 1	Mar. 7	12	comb + slight amt. of honey	0	-----
46	Feb. 29	Mar. 7	8	comb + slight amt. of honey	0	-----
47	Feb. 25	Mar. 7	6	honey + comb	0	-----
48	Mar. 8	Mar. 8	39	honey + comb	0	-----
49	Mar. 9	Mar. 10	21	comb + honey, pollen cells	0	-----
50	Mar. 21	Mar. 21	42	comb + honey, pollen cells	0	-----
51	Mar. 22	Mar. 22	33	comb + honey, pollen cells	0	-----
52	Mar. 22	Mar. 23	42	comb + honey, pollen cells	0	-----
53	Mar. 24	Mar. 24	21	comb, comb + honey, pollen cells	0	-----
54	Mar. 24	Mar. 25	18	honey + comb, pollen cells	0	-----
55	Mar. 25	Mar. 26	51	280 live bees	0	-----
56	Mar. 28	Mar. 29	21	110 live bees	0	-----
57	Apr. 19	Apr. 19	24	honey + comb, comb	0	-----
58	Apr. 19	Apr. 20	36	honey + comb	0	-----
59	Apr. 19	Apr. 21	36	honey + comb	0	-----
60	June 13	June 16	66	honey + comb, pollen cells	02	-----
61	June 28	June 29	60	honey + comb, pollen cells, 5 larvae	02	-----
62	July 14	July 21	90	honey + comb, comb	0	-----
63	July 25	Aug. 1	96	comb	0	-----
64	Aug. 5	Aug. 9	87	comb, comb + honey	0	-----
1933		1933				
65	Jan.	Jan. 12	45	comb, pollen cells, comb + honey	0	-----
66	Jan.	Feb. 11	30	pollen cells + slight honey	0	-----
67	Feb.	Feb. 16	33	crystallized honey + comb, pollen cells	0	-----
68	Feb.	Feb. 16	45	larvae + brood cells, crystallized honey + comb	0	-----
69	Feb.	Feb. 21	39	crystallized honey + comb, pollen cells	0	-----

²Two infections were obtained around point of inoculation in each series of Experiments 60 and 61, but as an uninoculated check also blighted, the infections in the inoculated shoots are of doubtful origin.

Table 5 shows that out of 3,534 injections, 21 resulted in infections with usual blight symptoms. In each of these infections (see Figures 15, 16, 17, and 18) the pathogen was isolated, grown in pure cultures from single colony transfers, reinoculated into healthy pear shoots, and typical infections again produced. All the material came from hives which were located in badly blighted apple and pear orchards, and it is of interest to note that the pathogen was obtained from material gathered in January, February, March, July, November, and December. With the exception of the series of experiments on a beehive and its

contents which was placed under pear trees completely enclosed with mosquito netting, no effort was made to obtain samples in the blooming period of pears or apples, that is, in the latter half of March and in the months of April and May. The reason for this exclusion was to avoid, if possible, the isolation of the organism shortly after it was introduced into the hive. The primary object of the experiments being to determine the longevity of the pathogen within beehives, there was no particular point in testing beehive material during and shortly after the blossoming period. If Gossard and Walton's data (6), not their conclusions, are accepted as a guide, one may expect a much larger percentage of successful isolations during the blossoming period. However, such isolations instead of representing overwintered inoculum, may just as readily be considered as of current spring introductions into the hive.

It is further to be noted in Table 5 that nearly all the successful isolations were made in 1929 and 1930, only one having been made in midsummer, after the 1930 blossom production, and none either before or after bloom production in 1932, or in the winter of 1932-1933. If such isolations were considered as indicative of the amount of contamination in beehives located in the apple-growing section of Arkansas during these years, is there any correlation between these findings and the amount of primary blossom blight which appeared in the respective years? According to these tests, 12 isolations having been made in the summer, winter, and early spring preceding the blossoming period of 1930, and 8 isolations in the winter preceding the 1931 bloom, the 1930 season, given favorable conditions for infections, should have been one in which an exceptional amount of blossom blight was to be expected, and 1931 should also have yielded considerable blossom blight, though with an appreciable reduction from 1930. Conversely, in accordance with these tests, 1932 and the coming season 1933 should be years in which primary blossom blight is very mild. How does this compare with the actual field data of blossom blight production? It has previously been noted that in 1930, blossom blight was extraordinarily abundant in spite of the total absence of rainfall during the whole of the blossoming period; in 1931 it was considerably reduced in quantity, though much more prevalent than in the succeeding year, 1932, when it was exceptionally low in amount. At the time this bulletin is being written (February, 1933) the amount of primary blossom blight for 1933 is unknown, but if these tests are at all valid it may be predicted that in 1933 there will be a relatively small amount of primary blossom blight in Arkansas, even if conditions are very favorable for infection.

Granting the validity of these tests, why is it that following

the extreme amount of blossom blight in 1930 there was not as great or greater amount of the same blight in 1931? In other words, if this form of blight is attributed to the oversummering and overwintering of the pathogen in beehives, it is to be expected that following a year of much blossom blight, there should be a considerable amount of inoculum available in the succeeding year. This may be expected to hold true provided environmental conditions within the hive are such as to permit the bacteria to remain alive and infectious. If unfavorable environmental factors intervene, such as extreme dryness or excessively high temperatures or an unusual abundance of microflora which is destructive to *Er. amylovora*,⁸ then in spite of large amounts of initial beehive contaminations the bacteria would not have the opportunity of remaining alive as long as in other seasons with more favorable environmental conditions but with a smaller initial number of bacteria. In this connection, the extraordinary dry hot summer of 1930 forces the consideration that under such conditions the life of *Er. amylovora* would very likely be much shorter than in seasons of normal rainfall and temperatures.

In addition to the possible deleterious action of adverse environmental conditions which may be operative when the bacteria are within the hive, there are other factors which may operate to prevent infections even in the presence of inoculum. Thus the degree of susceptibility of hosts, which varies with weather and soil conditions, and the fact that winds, cool weather, or rainy periods will hinder or prevent the activity of honeybees, must be considered in attempting to determine the factors which may prevent the occurrence of primary blossom blight.

Despite the care and relatively large amount of labor involved in attempting to find the fire-blight bacteria in beehives, the writer looks upon this work as suggestive but not conclusive. Considering the number of trials, the number of successful isolations is too small to be entirely reliable. It will require a great deal more work before this question is solved beyond reasonable doubt. Furthermore, even if beehives are found to be contaminated, this in itself is not sufficient evidence to substantiate the belief that bees coming from such hives are responsible for primary blossom blight. The recent report by Pierstorff (23) in which he failed to obtain blossom infections on apple trees which were in the presence of contaminated beehives, suggests caution in accepting this belief. As against Pierstorff's findings, however, there are a number of previous accounts of blight being absent on pear and apple trees prior to the introduction of bee-

⁸The writer with the help of Luther Shaw has found (28) that at least one microorganism often associated with fire blight has a destructive action on *Er. amylovora*.

hives, and following such introduction the disease appeared in short order. As a working hypothesis, the writer considers the beehive as a potential means of introducing and distributing blight in Arkansas, and for the present it is essential to take this into consideration in any program for disease control.

The following data briefly summarized, suggest the honeybee as a distributor of primary and secondary blossom blight in Arkansas, since this insect is the commonest blossom visitor of apple and pear blossoms in the state.

First, the absence of fresh, infectious exudate from overwintered blight prior to the appearance of first spring blight.

Second, the large percentage of cases in which blossom blight is the first form of the disease to appear.

Third, the advent of blossom blight in orchards in which blight was absent in the previous year.

Fourth, the absence of the disease on closed blossoms, and the finding of first blight symptoms in the upper parts of the flowers, suggesting disease initiation on the floral discs when blossoms are open.

Fifth, the great abundance of blossom blight in years of no rainfall and of warm sunny weather during the blossoming period, making conditions favorable for the activity of honeybees.

Sixth, the relative absence of blight on young or old non-flowering trees, particularly in the early part of the growing season, when nearby flowering trees are severely blighted.

Seventh, the isolation of the fire-blight pathogen from beehive material and from bees taken from hives in the winter and early spring prior to the appearance of new blight.

While the evidence presented above indicates that bees may often be involved in distributing primary and secondary blossom blight, this does not necessarily mean that all blossom blight is due to the activity of bees. Occasionally the writer has found bark beetles in association with both spur and twig blight. This, however, was relatively rare. In addition, he has observed instances where blossom blight having first appeared on upper limbs, spread during a period of rainy weather to numerous other blossoms and shoots located below and around the first infections. This has been especially noted in some of the seasons when the blossoming periods were greatly prolonged and when some of the first or central blossoms opened two to three weeks before the later ones. Similarly, later blooming varieties growing in the vicinity of others which blossom earlier, would likewise be subject to infection by means of rain-driven inoculum from the earlier blooming and earlier blighting varieties. While this has not been observed in Arkansas, Miller's report of such finding in southern Wisconsin (20), a region subject to cyclonic

storms similar to those occurring in Arkansas, suggests that this may also be expected here. On the other hand, Pierstorff's observations in southern Ohio (23) on the development of blossom blight on late-blooming apples, in which rain seemingly played no part in blight distribution and in which he attributed the dissemination to insect vectors, is not necessarily in conflict with Miller's observations. Both rain and pollinating insects are doubtless involved at times in distributing inoculum, particularly in those regions where rainfall is common in the blossoming periods. If any criticism can be made of the former work on means of fire-blight dissemination, it is that investigators as a whole are too likely to assume that the phenomena observed in some particular region will be the same as those occurring in other regions, and that the observations of a limited number of seasons will be comparable to those which may be expected at any time in a given locality.

INVESTIGATIONS ON OTHER INSECTS AS BLIGHT DISTRIBUTORS

In addition to bees, flies and wasps, at least 30 other species of insects have been suspected as being carriers and distributors of the fire-blight pathogen (7), including aphids, ants, leaf hoppers, tarnished plant bugs, bark beetles, wooly aphids, curculios, and mealy bugs. The literature has been reviewed so frequently in the past (20, 41, and 23) that there is no need of going into detail. In some instances, as for example bark beetles, (13) the evidence is quite substantial that they are contaminated with the fire-blight pathogen at times, and serve to distribute the disease. In most cases, however, investigators have relied to considerable degree on field observations relative to the incidence of insect infestations and the prevalence of blight. Rarely have efforts been made to determine the presence of the bacteria on or in the insect bodies, and in a few instances where this has been tried, the results were mainly negative.

While the writer has gathered a considerable body of field notes on the prevalence of various species of insects in blighted orchards, he is of the opinion that such data are difficult to evaluate in Arkansas because of the complicating factors of rainfall, heavy dews, and mists prevalent at times throughout the growing periods. Of course, in other sections of the country where meteoric water is extremely rare or absent during growing seasons, such data would perhaps have greater value. It may be recorded that in Arkansas leaf hoppers are more or less common on pears and apples, even during the blossoming periods, and that aphids are also present at times. Ants have frequently been noted traveling over diseased as well as healthy portions of pear and apple trees, and occasionally they have been observed localized in great abundance on apple trees bear-

ing diseased twigs and flower clusters. However, in no case was clear cut evidence observed of any general correlation of blight distribution and the prevalence of such insects. On the contrary, in some years, as in 1930 when the main outbreak of twig blight on apples appeared in late May after almost incessant rainfall throughout the first half of the month, neither leaf hoppers nor aphids were noted as abundant, and with such continuous rains plus low temperatures, it is obvious that insect activity especially of leaf hoppers would be considerably reduced.

Aside from field observations, the following lines of investigation were conducted to determine the role of various insect species as distributing agents or as agents which, by their wound-making ability, enable infections to be initiated: first, attempts at isolating the pathogen from insects; second, determining the degree of infectivity of various plant parts in the absence of insects; and third, determining the infectivity of plant organs in the presence of insects.

In the first line of these investigations, whenever an insect or group of insects was found in association with fresh blight, either in the field or in the greenhouse, the insects were handled with aseptic precautions, mashed in sterile water, and used for a series of poured plates and for injections into healthy pear shoots. The species of insects which were thus treated included mealy bugs, aphids (*Aphis pomi*?), and wooly aphids (*Eriosoma lanigera*). The number of these tests was very limited owing to the limited number of associations between blight and insects which were observed, and did not exceed six different trials. In each case the insects were found in direct association with freshly blighted plant parts, and the results are so contrary to prevailing opinions that they require further work. No successful isolations or infections were obtained.

In the second series of investigations an effort was made to produce infections on young leaves, succulent stems and various floral organs in the absence of any insects. This was repeated numerous times and always with the same results. In most instances it was attempted on potted pear trees and in a few cases with young, potted Jonathan apple trees. In all cases the trees were thoroughly inspected and if insects were present, they were removed either by hand or the trees sprayed with a solution of nicotine sulphate for the control of aphids. In each instance the inoculum was applied, by means of sterilized, metallic atomizers, as a spray consisting of pure cultures of *Er. amylovora* suspended in water. Immediately after the application of the inoculum and after a number of trees were sprayed with sterile water to serve as controls, all were placed in a cloth-enclosed, moist chamber located on a greenhouse bench, and left in the chamber for 48 to 72 hours. No record was kept of the

total number of infections produced, but it would run into the thousands. It has already been recorded that in one series of such inoculations, involving Kieffer pear blossoms and leaves, applied in the absence of insects, 196 separate infections were obtained and considering the fact that the controls invariably remained free from disease, the evidence appears quite conclusive. Considering the relative ease with which infections are obtainable on very young pear and apple leaves and succulent stems, without the aid of insects or of any observable wounds, it is almost unbelievable that for so many years the belief that infections on these parts require a wound, should have gone unchallenged.

However, from a reading of the most recent literature, including the very excellent second edition of the textbook by Heald (10), it does not appear that up to the present time the true significance of this discovery has been properly evaluated. To the writer, its importance rests not upon the explanation it offers for the entrance of the pathogen into unwounded leaves, but upon the fact that it explains the mode of entry of a large proportion of blossom blight, twig blight, and indirectly limb, body, and collar blight. Furthermore, by offering an explanation for twig infections in the absence of insects, it effectively reduces the need for the theory held for many years concerning the importance of the dissemination of the pathogen by insects, and the origin of twig blight by the action of contaminated and wound-inflicting insects.

To Heald (8) belongs the credit of offering the first observations, in 1915, relative to the penetration of fire-blight bacteria through the natural openings of pear and apple leaves, although the final proof for infection through natural openings did not appear until about 14 years later. In 1927 Heald (9, p. 350) reported artificial leaf infections through marginal breaks, insect punctures, or "through perfectly sound leaves". Then appeared in rapid order the work of Miller, Tullis, and Rosen, who presented irrefutable evidence for penetration of unwounded tissues. While the first published accounts of stomatal penetration by Miller (20) and Rosen (27) precede that of Tullis (41), it appears that the latter had initiated experiments and obtained excellent evidence for it about a year before the other investigators had started. Thus while Miller (20, p. 598, Table 7) apparently began to investigate twig infections in 1928, and Rosen (27, p. 69) in the same year, Tullis (41, p. 24, Table 6) obtained leaf infections in the absence of wounds in March, 1927. The writer's interest and approach to this question arose from the fact that in studying the host range of *Er. amylovora*, jointly with Groves (33), he was greatly impressed by the fact that unlike most bacterial species with wide ranges of hosts, this species could enter

the unwounded surface of at least one type of tissue, the nectary, whereas other species, such as *Er. carotovora* and *Phytomonas tumefaciens*, were apparently incapable of effecting entrance into any tissue in the absence of wounds. What prevents the entrance of *Er. amylovora* into any organ which possesses natural openings? It was to answer this question, rather than to find out how leaf blight or twig blight occurs in nature, that led the author to investigate invasions on various plant parts in the absence of wounds. Furthermore, the writer found, as previously noted, that despite the presence at times of aphids, mealy bugs, red spiders, and white flies in the experimental greenhouse, accidental contaminations on over 100 potted pear trees, which have been grown in it for the last 7 years, very rarely occurred, unless overhead watering was practiced. It is well known that even hydrocyanic acid fumigation will not easily control such insects as mealy bugs and red spiders without injuring the plants, although aphids are readily killed by it. While such fumigation has been employed in the greenhouse from time to time, unless other steps were taken to keep the plants free from insects, they were nearly always present to a greater or less degree. Why then should contaminations be restricted almost entirely to those periods following sprinkling of the tops, when one or more species of insects that are commonly supposed to act as blight distributors were nearly always present?

The question may be asked, why had so many able investigators failed to produce infections in the absence of wounds? The correct answer probably differs with different cases, but in many instances it is, in the light of the writer's work and that of Miller and Tullis very probable that many of the failures were due to an improper understanding of the relationship of age of host tissue to infection, the failure to grasp the importance of sufficient moisture within the tissues and on the surface, as conditioning agents for stomatal penetration, and the fact that wounds occurring naturally, such as growth rifts and cracks, hail injuries, and various mechanical abrasions caused by storms and winds and those that are man-made, such as scalpel wounds, needle pricks, pruning operations, are all conducive to infection, even of tissues which are otherwise resistant to stomatal invasion. All three of the investigators mentioned above, working independently of each other and in widely separated sections of the country, found that while very young pear and apple leaves are subject to infection through stomata, older leaves show marked resistance to stomatal penetration, although, according to Rosen (29, p. 56), they are still capable of being infected by means of wounds. Thus while wound infections may occur over a relatively wide range of time and conditions, stomatal infections are quite limited, although they are easily obtainable within those limits both under natural and artificial conditions. Evi-

dently, unless one understood the limits of stomatal penetration, the ability to produce infections through these natural openings would be more or less a matter of chance.

The writer's work clearly shows that in the absence of insects all that is necessary to obtain abundant leaf infections on both pears and apples is to apply a water-suspension of bacteria on unfolding bud leaves, the plants being kept under ordinary growing conditions with an air temperature around 25°C., with soil moisture sufficient to sustain continued growth of the leaves, and with the prevention of a rapid drying out of the sprayed plants.



Figure 19. Naturally infected Kieffer pear shoots, in which the infection in every instance was associated with abrasion of stem tissues. Material gathered near Bentonville, April 27, 1932.

TWIG INFECTIONS AS RELATED TO THE PRESENCE OR ABSENCE OF INSECTS

In a previous publication the writer (*29*, p. 52) noted that natural leaf infections are of three different kinds, those resulting from direct invasions through natural openings, those resulting from wound invasions, and those resulting from indirect invasion from stem to leaf by means of the petioles. The last kind was considered as being the most common. From the standpoint of twig infections, this means that the writer considered most twig

infections to have occurred first on the stems rather than on the leaves. This view has not been confirmed by some of the more recent publications. Both Tullis (41, p. 25) and Miller (20, p. 596-597) have decided that twig infections are largely traceable to infections on leaves rather than stems. Accordingly, a somewhat thorough study was made of twig infections under natural and artificial conditions on pear and apple.

When young, natural, twig infections are carefully observed, it is not difficult to find early infections which are limited entirely to the tip end of the stem, and to localized infections on succulent stems below the tip, both of which are entirely free from subtending leaf infections (see lower shoot and the one at the extreme left in Figure 19). Numerous blighted twigs are also to be found which show a blighting of both stems and leaves (see upper shoot and the one at the extreme right, Figure 19). In this last type, it is, of course, very difficult to tell whether the infections began on the stems or on the leaves, but there can be no doubt that in some instances it started on leaves and gradually reached the stem by means of midribs and petioles. However, according to the writer's artificial inoculation tests, stem infections which are initiated on leaves involve bud leaves largely in which infections occur in the absence of new stem-growth, when the developing leaves are borne directly on older wood. In these instances twig blight consists not of new shoot-growth but of one-year old stems. Such infections are readily produced artificially (see Figure 28, clusters of small leaves to the lower left) by means of water sprays of *Er. amylopora* in the absence of insects. In artificial water-spray inoculations, leaf infections which gradually involve new shoot growth are apparently produced in greater numbers than are usually found under natural conditions, (see Figure 20, upper left) and this may explain the conclusions of Miller and Tullis relative to stem infections being largely due to infections which originated on leaves. It is, of course, possible that in Wisconsin and Michigan such types of stem infections may be more common than in Arkansas, although the literature involving New York, Ohio, Washington, D. C., and other areas does not lend aid to this idea. The relatively few infections of this type which the writer has observed in apple and pear orchards were almost entirely restricted to the early part of the growing period.

A careful study of those twig infections which occur during the periods of maximum twig-blight development seemingly indicates that such blight in the greatest number of cases starts directly on the stems (see Figures 21, 22, 23, and 26). When the disease is in its initial stages, such stem infections are nearly always found to be free from infections which involve extensive leaf infections, as may be noted in the photographs. Indeed the



Figure 20. Artificial infections on Bartlett pear on both stem and leaves induced by watering in the vicinity of infected pear twigs, the watering being in the form of a spray from the nozzle of a hose attached to a water hydrant. No effort was made to keep the inoculum from drying-out after the application of the water-spray. Note infections on leaves, resulting in stem infection, in the upper left hand shoot, and on direct infections through wounds (leaf traces) made by removing the leaves.

evidence clearly indicates that infections which are initiated on stems, quite frequently pass up into leaf petioles, (see Figure 22, lower left), and may involve the base of the leaf blades, including midrib and adjoining laminar tissue.

The evidence for natural infections being initiated on stems, is fortified by artificial infections induced by water-spray inoculations of *Er. amylovora*. When such inoculations are made in the absence of insects, it is not uncommon to obtain stem infections as well as leaf infections (see Figures 27, 28, and 29), the two types being often independent of each other, and very comparable to infections found occurring in nature. Under natural conditions one may observe at the same time (1) more or less extensive twig blight involving stems and subtending leaves, (2) localized stem infections, either at the tips of the shoot or lower down, and (3) localized leaf infections either in the form of stomatal invasions of young leaves (see Figure 23, upper middle), or of relatively large-sized marginal invasions through wounds and perhaps hydathodes (see Figures 23, lower leaf, and 24).

Where and how are localized stem infections initiated? First, it is to be emphasized that they are not confined to the tips but may be found in greater or less abundance almost on any part of the current year's growth, between the nodes or immediately at the nodes. Frequently they may be observed starting in the axils of leaves (see Figures 24, 25, and 26 of natural infections; and 29, extreme right, of artificial infections). Since such infections have been produced artificially in the absence of insects or of observed wounds, it seems reasonable to assume that penetration has occurred through the natural openings, the stomata, of the young stems. On such stems the guard cells may still be observed (see Figure 30, lower right), and are evidently functioning in regulating stomatal openings, though in older stems they are often replaced by lenticel tissues which eventually disrupt the stomata and replace them by closing cells originating from lenticel phellogen. Why are initial stem infections more or less common in the axils of leaves? Is it because insects or wounds are more likely to occur in these somewhat secluded areas? Apparently not, judging by field evidence and by evidence obtained in artificial infections in the absence of insects. These axillary infections may perhaps best be explained by the fact that rainfall or water from mists and dews is more likely to cling and to remain for longer periods of time in these natural pockets than on exposed, internodal surfaces. When bacterial exudate or films reach these pockets of moisture, they are likely to remain in place and find suitable conditions for entrance into the tissues. These axils are not only lined with stomata on both petiolar base and stem, but are also the seat of

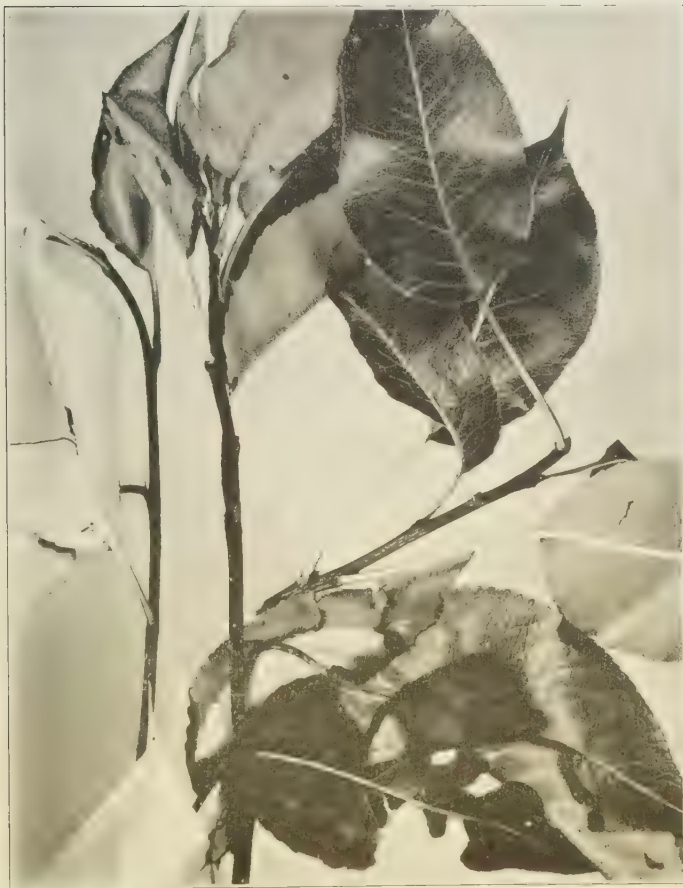


Figure 21. Natural infections of Kieffer pear shoots, involving stem infections wholly. Material gathered near Bentonville, on June 5, 1931

glandular hairs, such as Woodcock and Tullis (51) have described and the writer has confirmed. What role such hairs may play as infection courts remains unknown, but secreting as they do a sticky fluid they may offer either mechanical or chemical accessories by which bacteria adhere better to that region and perhaps are stimulated in growth and in activity.

The writer is by no means the first one to have discovered

axillary leaf infections on both pear and apple. Some 27 years ago Waite (48) reported, "Twigs (pear) are found with blight starting in the axils of the leaves or in the tender bark where no (insect) puncture can be found on careful examination." Stewart (38, p. 339) likewise noted and pictured bacterial invasions which "are frequently introduced into the tissues at the base of a petiole (Figure 117)", and in a later article jointly with Leonard (39, p. 158) he wrote, "Relatively few fire-blight infections which originate in the leaf ever extend down the petiole to the twig."

In order to gain further information on stem infections which are independent of leaf invasions, the leaves were stripped from several potted Bartlett pear trees, sprayed with a water suspension of the fire-blight pathogen, a similar number of denuded trees were sprayed with sterile water to serve as controls, and all placed in a moist chamber for 48 hours. No infections appeared on the controls. On the trees which received the bacterial applications, numerous infections were noted, (see Fig. 30), many of them centering or beginning in and around the leaf traces, and some of them, in the form of much smaller, localized diseased areas, centering around stomata and young lenticels (see Figure 30, right). Nearly every leaf-trace region became infected, the disease developing very rapidly and involving considerable stem tissue as may be seen in Figure 30. In contrast to these, the stomatal invasions in most instances resulted in more restricted infections. While there can be no doubt that the wounded stem regions around the leaf traces offered much superior avenues for infection, the evidence for infection through the natural openings on the stem is fairly adequate. Nevertheless, considering the fact that these experiments involved the use of relatively large amounts of pure culture inoculum, which with the possible exception of twigs located close to heavy bacterial exudations would be greater than may be expected to be deposited in nature, it appears that wounds on succulent stems offer greater possibilities as infection courts than natural openings.

These experiments on wound infection through freshly exposed leaf traces do not, however, give support to the assumption that insects are largely responsible for twig infection. No insects were known to be present in these trials. Furthermore, the large wounds resulting from tearing leaf petioles from stems bear no resemblance to the relatively minute wounds occasioned by stem-feeding insects such as aphids, tarnished plant bugs, mealy bugs, and wooly aphids. In fact it may be questioned whether insects with sucking mouth parts, such as aphids, produce open wounds, the very few epidermal cells injured by the fine piercing setae representing areas which are of microscopic proportions and hardly in the nature of naked wounds. Leaf hoppers



Figure 22. Natural infections on Kieffer pear shoots involving stem infections, and showing the passage of the disease from stem to petioles (lower left-hand shoot). Material gathered near Bentonville, on June 11, 1931



Figure 23. Natural leaf and stem infections on Kieffer pear. Note stem infections independent of leaf infections, and two types of leaf invasion, one-stomatal (upper middle), and two-marginal, presumably through a tear (lowermost leaf). Material gathered near Bentonville on June 5, 1931.

being almost entirely confined to lower leaf surfaces would in the light of the investigations here reported, have little to do with twig blight as a whole. Likewise, bark beetles which undoubtedly are responsible for blight dissemination at times, are not sufficiently abundant on healthy, vigorous-growing trees to account for the widespread occurrence of twig blight. It is of course well known that these insects feed mainly on weak-growing, debilitated or partly dead limbs. Pear thrips are not known to occur in Arkansas, but wherever they are present in abundance, as in the far western states, it is possible that the type of injury which they produce would be conducive to blight initiation. From the standpoint of size and nature of wound in relation to blight production, chewing and biting insects such as canker worms and caterpillars, are far more likely to induce blight than sucking insects. Fortunately, chewing and biting insects are not ordinarily present in abundance in Arkansas during the periods of blight development.

As a further aid in determining the role of sucking insects as vectors, a number of potted pear trees which became infested with aphids and upon which the insects were intentionally permitted to multiply, were inoculated with a water spray of *Er. amylovora* and placed in a moist chamber for 48 hours. At the same time a group of insect-free trees were similarly treated and placed next to the insect-infested ones. As a further check, another group of trees that were free from insects were sprayed with sterile water and placed in the moist chamber. The last group developed no blight. The insect-free group sprayed with the fire-blight pathogen developed a number of blight lesions on both stems and leaves, the diseased stem areas being in some instances very pronounced (see Figure 29). The trees infested with aphids, the infestations being so heavy that leaf curling and stem dwarfing were noticeable (see Figure 29 on the left), showed a very much reduced amount of blight, both in number and in size of lesions, compared with the insect-free trees (see Figure 29). There is very little doubt that in this experiment the insects instead of aiding blight-development actually retarded it. This perhaps is to be expected, since anything which serves to hinder growth is likely to retard or prevent fire blight.

The fact that many natural infections and those induced artificially can be traced to natural openings, and the fact that sucking insects of the types previously mentioned seemingly play a minor role in disease dissemination, does not mean that wounds are unimportant avenues for infection. Under field conditions the writer has observed numerous infections on both stems and leaves which were seemingly traceable to wounds, not insect wounds in most instances but those produced by strong winds, rain storms, and hail injury. Not infrequently infections were noted starting on portions of stems that had suffered abras-



Figure 24. Natural infections around axils of leaves and on leaf blades of Kieffer pear. Material gathered near Bentonville on June 11, 1931.

ion by rubbing against other tree parts. Likewise, infections have often been observed occurring around wounds when infections traceable to natural openings were either absent or undetected. This has frequently been the case in infections which were initiated in the latter half of June and in July, when stomatal infections were extremely rare. However, it is not uncommon to find wound infections at any time during the first half of the growing season, and as far as leaves are concerned, it is perhaps true that the greatest number of infections are initiated in



Figure 25. Natural infections initiated on stems or leaf axils on Kieffer pear. Material gathered at Bentonville, on June 5, 1931

broken or cracked areas. This is certainly the case in full grown leaves. While there can be no question about bacterial penetration through natural openings on both leaves and stems, there also is no doubt that mechanical wounds play a very important part as disease portals in Arkansas, and probably in other states.

The evidence here presented points to the conclusion that control measures which aim at eliminating certain sucking insects (aphids and leaf hoppers) by the application of nicotine sulphate, lime sulphur, or other contact insecticides would have little value as blight preventives. On the other hand, the application of germicidal sprays to prevent infections through natural openings in the early part of the growing season in Arkansas, a spray which will cover floral parts, stems, and leaves, is suggested by the findings that have been presented here. That such sprays cannot be expected to control blight at all times is also predictable on the basis that they could not prevent infections through mechanical wounds, especially those that are made during stormy periods, and through certain insect wounds, such as those made by thrips and bark beetles.

ATTEMPTS TO CONTROL FIRE BLIGHT BY MEANS OF SPRAYS

For many years investigators and growers have claimed at times that spray applications of Bordeaux mixture and other fungicides will control blight. In 1892 Waite (*42*) reported that "trees infected with blight germs artificially may be sprayed with fungicides and the greater part of the damage prevented," and in another publication issued in the same year (*43*) he states that a 3 per cent solution of chloride of lime "gave absolutely perfect results, but scorched the foliage slightly. Bordeaux mixture and ammoniacal solution of copper carbonate were the other fungicides used, and gave good results." In his later publications (*44, 45, 46, 47, and 48*), however, he no longer recommended the use of fungicidal sprays, and reported in 1906, "As a rule spraying is of little use, as I have tried repeatedly, by using Bordeaux mixture and other compounds."

In 1901 Chester (*3*) in Delaware recommended spraying with Bordeaux mixture, using it also as a wash for trunks for the prevention of blight. In 1909 (not reported until 1917) McCue (*19*) working in the same state found that 5 Bartlett pear trees sprayed with Bordeaux mixture when the trees were in full bloom, showed a considerable reduction in blossom blight.

In 1917 Stevens and his associates (*36*) working in Illinois, reported that Bordeaux mixture controlled floral infections without reducing the set of fruit.

Reimer in Oregon (*24*) in 1926 and in subsequent years (*25, 26*) recommended spraying with Bordeaux mixture as one of the



Figure 26. Natural infections on leaf blades and in axils of leaves of Kieffer pear. Note particularly the axillary infection in the lower-center position. Material gathered at Bentonville, June 5, 1931

measures to be used in blight control on pears on the theory that this material in addition to having powerful germicidal properties acts as a repellant for crawling insects. He began a series of spray experiments with this material in 1923 and has continued it since then. The mixture, made up of 3 pounds of copper sulphate, 6 pounds of lime, and 50 gallons of water, was applied during the pink stage and immediately after petal-fall. "The results showed a very material reduction in the number of blight infections where the spray was applied." One year when the first application was delayed until the trees were in full bloom, no benefit was derived from the spraying. In 1930 (26) he stated further that, as a result of these experiments, spraying with Bordeaux had become a well established practice in some of the commercial orchards in the Rogue River Valley with beneficial results.

In 1927 Morrison (21) in an address presented before the Placer County (California) fruit growers association, reported that some pear growers in Sacramento County who had followed his instructions in applying Bordeaux sprays, had succeeded in controlling blight perfectly, whereas others experienced a total failure. From the notes presented of the different spray tests in 1926 and 1927, it appears that some growers used only one spray applied with arsenate of lead, apparently after petal fall, others used two sprays, the time of application varying in different orchards. In one instance it was presumably applied in the "pink stage" (April 11, 1927) and after petal fall; in others, both applications were made after petal fall, presumably when the usual codling-moth sprays were applied. No correlation was noted between the number or time of applications and the amount of blight developed. The different growers using the sprays at different times, evidently intended to determine by the "hit or miss" system, the effectiveness of Bordeaux sprays for blight control.

McCown (17, 18) working in Indiana in 1927 and 1928, found that when Bordeaux mixture of a 1-3-50 formula was applied to open apple blossoms and was followed by an application of pure culture inoculum, there was a considerable reduction in the number of infections, particularly when the bacteria were applied immediately after the Bordeaux spray. In those inoculations made 24 and 48 hours after the spray application, the percentage of infected clusters was considerable, although somewhat less than the unsprayed and inoculated checks. He also applied one spray of 1-3-50 Bordeaux mixture on 14 apple trees, 10 in one orchard and four in another, and on three pear trees, when the trees were at or near full bloom, and reports considerable reduction in the number of infections compared with unsprayed checks. McCown (17, p. 6) believes that under Indiana condi-



Figure 27. Artificial stem and leaf infections induced by water-spray inoculations of *Er. amylovora*. Plants kept in a moist chamber 48 hours after inoculation. Note stem infection on right-hand shoot which is independent of those on leaf-blades. Infections produced in the absence of insects. Photographed July, 1931.

tions, rain is the most important distributing agent of the bacteria from holdover canker to blossom, and if a germicidal spray is applied to open blossoms before a rain and before pollinating insects begin to visit the blossoms, blossom blight and to a considerable degree twig and limb blight, will be controlled.

Contrary to McCown's findings in Indiana, Miller in Wisconsin (20, p. 614) found that a single blossom-spray of Bordeaux mixture had little or no influence in controlling blossom blight, although he applied this mixture 2 years in succession, 1926 and 1927, and sprayed 140 trees the first year and 80 trees the next year. On the other hand, Keitt, Shaw, and Riker (14, 15) reported in 1932 that a single full blossom spray applied to large apple trees, upon which bacterial inoculum was applied artificially, gave promising results in blight control in Wisconsin.

In Tennessee, Sherbakoff in 1931 (35) applied 1-3-50 Bordeaux mixture to Yellow Transparent apples, eight trees being sprayed once when early in blossom, eight others once late in blossom, and eight twice, early and late in blossom. "The results were somewhat encouraging, but gave no great promise." The trees sprayed twice during the blossoming period yielded 46 per cent blight of the blossom clusters, while 13 unsprayed trees averaged 68 per cent blossom blight.

From a review of the literature on Bordeaux-spray applications presented above, it will be seen that for every report of success in controlling blight by this method, there are one or more reporting failure. It is likewise obvious that the tests in most instances were undertaken with the hope of finding some suitable control even though the fundamental biological principles concerning the manner of overwintering of the pathogen, the time of inoculum dissemination, and the agents involved in distributing the bacteria were unknown for the region in which the investigator was concerned. Under such conditions it is not at all surprising that the reported results are so diverse.

It is obvious that sprays to be effective must be applied at the time when the various sources of inoculum for primary and secondary blight are operative. Evidence has been presented to show that under Arkansas conditions there are a number of possible sources of primary blight, including: first, contaminated beehives, in which the bees may carry the bacteria from the hive to open blossoms; second, internal extension of blight which results in bacterial exudations during the latter part of the blooming season and during the production of early shoot growth; third, infected buds which may result in twig blight on the older wood or on the new shoots arising from the buds; fourth, masses of bacteria representing exudates of the previous season remaining alive on the surface of contaminated tree-areas; and fifth, overwintered limb blight which produces bacterial exudates in



Figure 28. Aerial infection on Bartlett pear induced by pure culture inoculations of *E. amylovora* after 10 days. The infection is shown as a water spray in the presence of aphids shoot to the surface of the leaves as a water spray. In the absence of insects, and excluding light heat, as a function by means of a hypodermic needle. A water spray is visible on the surface of the leaves. Photographed 3 days after inoculation.

late May and early June. Clearly in any orchard where twig and limb blight is more or less common, despite excision practices, one or two sprays cannot be expected to yield effective control, although the amount of infection may be somewhat reduced. On the other hand, in any orchard where blight is largely confined to blossom clusters or usually present in the form of small amounts of twig blight, two sprays applied during the blossoming period may control blight very effectively, particularly in those orchards which are not adjacent to or in the immediate vicinity of badly blighted trees.

It is also obvious from the evidence previously presented, that considering the marked differences in climatic conditions and the differences in varieties grown in different sections of the country, the time of inoculum production, particularly the exudations from overwintered blight, may be expected to appear at unlike periods of growth in the different sections. Thus in Wisconsin, and possibly other northern states east of the Rockies where the evidence is excellent for exudations appearing very early from overwintered blight, it would seem that one or two sprays, comparable in time of application to the pre-pink and cluster-bud sprays for apple scab, should be effective in controlling that form of blight attributable to overwintered cankers. And if this is combined with a full-bloom spray to serve as a protection against contaminated, pollinating insects which may bring in secondary blight from early blooming pomes, such as crab apples, blight control may be more certain than it is at present. On the other hand, in other sections of the country, as in Arkansas and comparable regions where exudations from overwintered blight do not appear until later in the growing season and where the bacteria may overwinter in beehives, the sprays to be effective must be applied at other times. Furthermore, when an orchard develops blight in the early part of the growing season, given favorable weather conditions for blight dissemination, such as rains accompanied by wound-inflicting winds or hail, spraying cannot be considered as offering much protection in the light of the investigations here presented.

An attempt having been made to determine the underlying biological reactions concerned in the overwintering and dissemination of the blight bacteria in Arkansas, the next step was to utilize these data in possible control measures. Having found that pruning of diseased wood, the chief control measure which has been utilized up to the present, very frequently does not protect the trees adequately, it appeared that the application of sprays which possess germicidal properties applied at the time when the bacteria are disseminated may possibly be of value as a preventive measure. And as Bordeaux mixture had frequently been mentioned in the literature as having exercised control



Figure 29. Artificial infections on aphid-free and aphid-infested Baretlett pear shoots induced by application with water-sprays of *Er. amylovora*, photographed 4 days after inoculations were made. The two shoots at the left were aphid-infested, those at the right were free from insects. Note absence of stem infections on the former and the marked stem infections of the latter.

over blight, it seemed desirable to give this material a thorough trial. The fact that numerous investigators had reported failure to control blight by the use of this material did not warrant the conclusion that it was ineffective, since many of these failures could be attributed to the lack of information on the sources of the bacterial inoculum and the time and agents involved in the distribution of the bacteria.

As the disease was in most instances found appearing first on open blossoms of both pear and apple, it seemed evident that the first spray to be effective must be applied in time to protect the blossoms from infection. Consequently, beginning with 1929 and continuing up to the present, the writer, with the help of an orchardist near Bentonville in Benton County, and another near Farmington, Washington County, has been applying a weak Bordeaux mixture consisting of one pound of copper sulphate, three pounds of hydrated lime, and 50 gallons of water, to 125 Jonathan trees when they were in bloom. However, up to 1932 the check trees adjacent to the trees sprayed with Bordeaux, did not develop sufficient blight to enable one to draw any conclusion relative to the effectiveness of Bordeaux mixture.

In 1932, the isolated apple orchard located on a mountain side near Ozark, previously described as having shown 95 per cent blossom blight in 1930 and 60 per cent in 1931 in a block of Jonathans, was utilized for a thorough spraying test. This block consisted of 167 trees, 22 years old. Four rows, comprising 64 trees, served as controls, receiving the regular early season spray applications common in many of the apple sections of America, consisting of a cluster bud or pink spray, a calyx spray and a first cover spray, the material being one and one-half gallons of commercial lime sulphur to 50 gallons of water, to which was added one and one-half pounds of arsenate of lead in the calyx spray and in the first cover spray. Seven rows, comprising 103 trees, growing adjacent to the check trees, received the experimental spray applications for the control of blossom blight. The material consisted of a weak Bordeaux mixture, made up of one pound of powdered copper sulphate, three pounds of hydrated lime, and 50 gallons of water. To this was added arsenate of lead in the calyx and cover sprays in the same amount used for the checks. The applications were made as follows: First, as a cluster bud or pink spray (April 8 and 9); second, when approximately 25 per cent of the blossoms were fully open (April 12); third, when approximately 80 per cent of the blossoms were open (April 16); fourth, when about seven-eighths of the petals had fallen (April 20); and fifth (May 9), when the first brood of codling moth was anticipated.

It is to be noted that, aside from chemicals, the main point of difference between the experimental spray program and that



Figure 30. Left—stem infections on Bartlett pear induced by water-suspensions of *Er. amulovora* in regions of wounds (leaf traces) and apparently through stomata or young lenticels. Upper right—surface view of an infection centering around one of the stomata. Magnified 180 X. Lower right—cross (radial) section of succulent stem of Bartlett pear at an infected area, the infection centering around a stomata. Infection produced by a spray of *Er. amylovora* suspended in sterile water. In the process of sectioning and mounting the infected material in water, the bacteria floated out of the material in huge numbers, and consequently are not observable. Note the marked discoloration of tissues centering around the stomata, and the absence of disease symptoms in the deeper tissues. Magnified 410

of the standard spray schedule is the application of two sprays when the blossoms were fully open. The danger of injury from spraying such delicate organs was present, of course, in the mind of the owner and of the investigator, but the latter's previous experience in applying one spray of Bordeaux mixture to open apple blossoms in other orchards for three successive years previous to this experiment, suggested that two such sprays would not result in injury to blossoms or in excessive russetting of fruit. It is well known that the latter type of injury, depending on weather conditions, may be had with the regular lime sulphur sprays as with Bordeaux mixture, although most observers agree that during cool, moist weather Bordeaux is likely to cause more russetting than lime sulphur. However, the writer's work up to the present suggests that in the Ozarks of Arkansas early season applications of weak Bordeaux on Jonathan apples, the only variety investigated, have not caused any large amount of injury to fruit and from this point of view have been satisfactory to the grower in every instance. The necessity of two spray applications to open blossoms is obvious. Very rarely will any large proportion of the blossoms open at the same time on any pome; often as much as 2 weeks or more intervenes in Arkansas between the opening of the first blossoms and the later ones.

The observations and results of this particular experiment to control blight may be briefly recorded as follows:

First, while much of the blighted wood of the previous year had been pruned out, numerous blighted twigs remained. Nevertheless, no exuding cankers were found on any of the trees prior to the first signs of blight.

Second, blight was first noted on April 25 in 22 blossom clusters on check trees. None was found on the Bordeaux sprayed trees. A thorough search on every tree for active hold-over cankers resulted in failure.

Third, blight was found in greater or less abundance on May 5 on almost every check tree, while the Bordeaux sprayed trees remained without any signs of blight. However, the number of blighted blossom clusters on these checks was as a whole not nearly as great as in the past few years. Only one check tree showed as much as 308 blighted clusters, or 60 per cent.

Fourth, diseased blossom clusters having turned brown by May 9, the disease was now more easily detected. Accurate counts were made again on each blighted tree. The amount of disease on the checks appeared to be the same as that noted on May 5. On the Bordeaux sprayed trees, only three of the 103 trees showed any blight, the total for the three being five blighted blossom clusters.

Fifth, on May 18, when many of the fruits had attained the size of one inch in diameter, secondary blight was noticeable on

about one-third of the check trees, though not in great quantities, and on two of the Bordeaux-sprayed trees standing in the row next to the checks. Four blighted leaf shoots comprised the total number of secondary infections on the Bordeaux-sprayed trees, a number which represented a very small fraction of the total number, uncounted, on the checks. These secondary infections occurred on leaf shoots devoid of fruit clusters as well as on twigs bearing both succulent shoots and fruit clusters. No further blight development occurred in this orchard for the remainder of this season.

The results of this experiment are so clear cut that there can be no doubt about blossom blight having been almost completely controlled in the experimental spray plot in 1932. Likewise, twig blight was also materially reduced. Viewed from a background of recurrent and disastrous epidemics of blight on pears and apples experienced in America for over a century, the results here cited are almost too good to be true. It appears doubtful to the writer that such nearly perfect control will always be obtained by this schedule. The results of this test, however, appear unmistakable. There remains to be determined the influence of this schedule on russetting of various varieties of pears and apples; its influence on setting and dropping of fruit; its effect on foliage, twig and limb growth; and its efficiency in controlling twig, limb, collar, and root blight. It is also possible that it may be effective in some years and in some sections of the country and not in others.

It has previously been shown that the form of blight common in this orchard over a period of years was predominantly blossom blight, with good indications that the inoculum was brought in by pollinating insects. In 1932, as in previous years, twig blight which was not in the immediate vicinity of blossom blight was extremely rare, being in most instances merely internal extensions of infections generated in the blossoms. The form of twig and limb blight which usually appears in late May and in June and which is commonly associated with overwintered cankers was wholly absent in this orchard. It was, therefore, unnecessary in this orchard to employ later sprays which might be employed against this source of inoculum. It is obvious, therefore, that in those orchards which possess hold over cankers on limbs of an inch or more in diameter, the above schedule cannot be expected to control the later or chief outbreak of twig and limb blight. Indeed, considering the fact that the cover sprays primarily designed for codling moth control and usually applied as a combination insecticide and fungicide spray, with 3-4-50 or 4-4-50 Bordeaux mixture often used for blotch and bitter rot control, has not resulted in any noticeable reduction in twig and limb blight, suggests that Bordeaux sprays aimed pri-

marily for controlling this form of blight may not be very effective. This, however, remains to be determined.

It seems desirable to point out that the five Bordeaux sprays applied as indicated above may be expected to control not only blossom blight, but should result in more effective control of leaf spot and fruit rot caused by *Phylospora cydoniae*, blotch, and to some extent bitter rot. On the other hand, powdery mildew, a disease which is relatively rare in Arkansas and in most of the apple-growing sections east of the Rockies, and scab, which is as a whole not nearly as severe on the blight-susceptible varieties grown in Arkansas as on blight-resistant ones, may not be expected to be controlled as well by these sprays as by the standard lime-sulphur treatments.

As to the influence of these Bordeaux sprays on appearance of foliage, on setting of fruit, and on spray injury to fruit, the following notes were taken: As a whole the trees sprayed with Bordeaux mixture in comparison with the adjoining trees sprayed with lime sulphur, showed a noticeable superiority in color and general vigor of the foliage. The leaves were darker green and persisted longer on the trees. The set of fruit did not seem to be materially affected, despite the two additional sprays of Bordeaux applied to the open blossoms. As to the amount of spray injury, the 103 trees sprayed five times with Bordeaux showed russetting to the extent of reducing the grade of 2 per cent of the fruit, contrasted with the fruit burning caused by the three lime sulphur sprays which reduced the grade of 3 per cent of the fruit. However, the finish on the fruit sprayed with lime sulphur was as a whole somewhat superior to that of the Bordeaux sprayed fruit. Considering the much more conspicuous type of spray injury induced by lime sulphur and the greater percentage of fruit reduced in quality, it would appear that despite the finer finish of the fruit which received lime-sulphur sprays, and irrespective of blight control, the Bordeaux-sprayed fruit had a greater marketable value than the lime-sulphur treated fruit, as far as the 1932 season was concerned. It is probable, however, that depending on weather conditions, the amount of spray injury induced by both kinds of sprays will vary from year to year and there are numerous references in the literature which suggest that in more northern regions Bordeaux spraying may be attended by greater amounts of fruit injury than lime-sulphur sprays. For the present the writer considers it to be ill-advised to eliminate lime-sulphur sprays in a wholesale fashion. In those orchards which usually remain free from blight or which develop blight in insignificant amounts, there is no good reason for using Bordeaux mixture any time up to the first cover spray. On the other hand, in those blocks of susceptible varieties where blight is more or less common and often very destruc-

tive. Bordeaux spraying gives promise of yielding outstanding results. However, even in these instances, growers are advised not to eliminate the lime-sulphur sprays entirely, until the results of several additional seasons are at hand. It would be desirable, however, for orchardists in general to apply these sprays to a limited number of trees of susceptible varieties in order that the material be tested as thoroughly and in as many localities as possible. The present prices of these materials are such that five sprays of Bordeaux mixture are somewhat less expensive than three sprays of commercial lime sulphur, copper sulphate being quoted at 4½ cents a pound in crystal form and 5 cents a pound in powdered form (the latter being preferable), hydrated lime of a good grade at 1 cent a pound, in contrast with commercial lime sulphur which is quoted at 10 cents a gallon.

SUMMARY AND CONCLUSIONS

Prior to the appearance of new infections, no bacterial exudations containing infectious bacteria were found issuing from overwintered blight during the months of February, March, and April for the years 1927 to 1932, inclusive. This study involved field observations of over 3,000 acres of susceptible apple varieties located in Washington and Benton counties of Arkansas.

Attention is directed to the marked differences in climatic conditions existing in Arkansas compared with more northern apple-growing regions, and the possible influence of diverse climatic factors on the disparate production of bacterial exudate. A theory is presented to explain the extrusion of bacteria in the form of exudates from blighted tissues.

In a discussion of pruning practices in Arkansas as related to blight control, it is noted that if the currently accepted theory relative to the control of blight by pruning is correct, then in those orchards which had little or no blight in the previous season and in those where pruning has removed most of the blighted branches, the pathogen is not likely to be present in the following season. Hence, in these instances fire blight may not be expected to appear unless the pathogen were brought into the orchard from an outside source, or unless overwintering, contrary to the current theory, is accomplished in some additional ways.

A study of the succession of blight in consecutive years in 92 apple orchards revealed numerous instances in which no correlation existed in one orchard or in a group of contiguous orchards between the presence or absence or amount of blight in one year compared with the succeeding year. In many cases blight in considerable quantities was found in 1930 in well-isolated orchards which had no blight in 1929 and which apparently were out of reach of rain-driven inoculum.

In attempting to determine the role of meteoric water as a conditioning agent for blight infection and as a distributing agent for blight bacteria, a study was made of blight development in relationship to weather conditions over a series of years. It is concluded that rainfall acts at times as an important agent of distribution in Arkansas and also probably as an important conditioning agent, directly and indirectly. In some years, however, as in 1930, a year in which blossom blight of apples was extremely prevalent and destructive in Arkansas, no rains having fallen throughout the period of blossom development, the deduction is made that instead of meteoric water acting as the agent of dissemination for this blight, pollinating insects were probably involved in large measure. On the other hand, the severe twig blight which developed later in that season, was apparently correlated with exceptional amounts of rainfall.

In studying the sequence of fire blight within a growing season, blossoms of both pears and apples were noted as being usually the first organs to show signs of infection. Artificial inoculations in large numbers were made both under field conditions and in the greenhouse in order to determine if this first form of blight may not be due to a difference in susceptibility of floral structures as compared with leaves. These experiments showed, first, that at the time of blossom production young leaves located either in the blossom cluster or arising from strictly vegetative buds are fully as susceptible to infection as any floral tissues; second, infections on flowers and leaves are readily obtainable in the absence of insects or wounds provided the inoculum is applied as a water-spray simulating rainfall or rain driven by wind.

When earliest symptoms of fire blight on pear blossoms are not complicated by frost injury, it is noted that the disease appears first in the upper parts of the flowers, suggesting that the pathogen was distributed by blossom-visiting insects and not by rain. Evidence is also presented which shows that pome blossoms may become infected, but when frost injury occurs blight symptoms are wanting, though droplets of bacterial exudate are produced, and abscission of such blossoms may occur.

It is concluded that in by far the greatest number of cases the first signs of blight are restricted to blossoms when frosts or extremely low temperatures do not intervene, otherwise this disease may appear simultaneously on blossoms and leafy shoots.

A study of twig blight on pears and apples indicates that, although much of it is secondary, being directly or indirectly connected with blossom blight, there is another type which shows no such connection. The latter, apparently of the nature of primary blight, may originate in one of four different ways: first, as internal extensions of the previous year's blight, which usually appears shortly after blossom blight has been evidenced; second, as bud-infections in which the buds, though infected the previous year by internal blight extensions, remain alive through the winter; third, as infections resulting from bacterial exudates produced in the previous growing season; and fourth, as the well-known twig blight induced by inoculum from overwintered cankers, a form of blight which in Arkansas, does not ordinarily appear until late May or in June.

In recording the details concerning the investigations on the possible role of honeybees as distributors of primary and secondary blossom blight, evidence is presented for the overwintering and overwintering of the fire-blight pathogen in the beehive at times, and further data given which seemingly connect blossom infections with the visitation of contaminated bees.

Studies on other insects as possible blight-disseminating

agents are reported and investigations concerning the commonly accepted theory relative to the necessity of wounds for the production of twig and leaf infections revealed that infections on various floral tissues, on leaves, and on shoots can be easily produced in the absence of insects or of wounds.

In studying the mode of entrance into succulent twigs and the production of twig blight under natural and artificial conditions, it is concluded that under natural conditions in Arkansas twig blight originates mainly on stems rather than on leaves. The entrance is seemingly effected in some instances through direct stomatal penetration of stems, often in the stem-regions located in the axils of leaves. However, experiments and field observations indicate that wounds offer avenues for disease inception on stems superior to those offered by natural openings. The type of wound found conducive to blight development is noted as being very unlike those produced by sucking insects such as aphids, and it is concluded that mechanical wounds play a very important part as disease portals on both stems and leaves.

The results obtained by the use of Bordeaux sprays in attempting to control blossom blight appear to offer considerable promise for better protection against this form of the disease in Arkansas.

LIST OF REFERENCES

- (1) Becker, G. G. Non-pear zones and blight eradication. Jour. Econ. Ent.
21: 485-487. 1928.
- (2) Brooks, A. N. Studies of the epidemiology and control of fire blight of
apple. Phytopathology 16: 665-696. Illus. 1926.
- (3) Chester, F. D. Pear blight and pear canker. Delaware Agr. Exp. Sta.
Bul. 52, 8 pp. Illus. 1901.
- (4) Cunningham, G. H. Fire blight and its control. New Zealand Dept. Agr.
Bul. 153. 8 pp. Illus. 1931.
- (5) Frisch, K. von. Aus dem Leben der Bienen. Berlin, 149 pp. Illus. 1927.
- (6) Gossard, H. A. and Walton, R. C. Dissemination of fire blight. Ohio
Agr. Exp. Sta. Bul. 357: 81-126. Illus. 1922.
- (7) Groves, A. B. A comprehensive bibliographical résumé of the disease
known as fire blight of apples and pears. Thesis, University of Ar-
kansas, 105 pp. 1928.
- (8) Heald, F. D. Preliminary note on leaf invasions by *Bacillus amylovorus*.
Washington Agr. Exp. Sta. Bul. 125, 7 pp. Illus. 1915.
- (9) ——— Leaf invasions by *Bacillus amylovorus*. Northwest
Science 1: 76-79. 1927.
- (10) ——— Manual of plant diseases. McGraw-Hill Book Co., New
York, Second Edition, 953 pp. Illus. 1933.
- (11) Hesler, L. R. Fire blight of fruit trees and its control. Proc. Ann. Conv.
Tennessee State Hort. Soc. 18: 51-57. Illus. 1923.
- (12) Hewitt, J. L. Twig blight and blossom blight of the apple. Ark. Agr.
Exp. Sta. Bul. 113: 493-505. 1913.
- (13) Jones, D. H. Bacterial blight of apple, pear, and quince trees. Ontario
(Canada) Agr. Coll. Bul. 176, 64 pp. Illus. 1909.
- (14) Keitt, G. W., Shaw, L., and Riker, A. J. Bactericides in relation to *Bacil-*
lus amylovorus and fire-blight control. Phytopathology 22: 15 (ab-
stract). 1932.
- (15) ——— ——— ——— Bordeaux gives
promise of checking fire blight. Wisconsin Agr. Exp. Sta. Bul. 421:
70-71. 1932.
- (16) McClarty, H. R. Fire blight. *Bacillus amylovorus* (Burr.) Trev. Rept.
Dominion (Canada) Botanist for 1922: 64-66. 1923.
- (17) McCown, M. Spraying for the control of fire blight in the apple. Trans.
Indiana Hort. Soc. 1927: 129-133. 1928.

- (18) ————— Bordeaux spray in the control of fire blight of apple. *Phytopathology* 19: 285-293. 1929.
- (19) McCue, C. A. Pear blight. *Trans. Peninsula Hort. Soc.* 1917: 51-55. 1917.
- (20) Miller, P. W. Studies of fire blight of apple in Wisconsin. *Jour. Agr. Res.* 39: 579-621. *Illus.* 1929.
- (21) Morrison, A. E. Pear blight control, Sacramento River District. Placer County (California) Fruit Growers' Convention 7th (1927): 35-38. 1928.
- (22) Owens, C. E. Principles of plant pathology. John Wiley & Sons, New York. 629 pp. *Illus.* 1928.
- (23) Pierstorff, A. L. Studies on the fire-blight organism, *Bacillus amylovorus*. N. Y. (Cornell) Agr. Exp. Sta. Memoir 136: 53 pp. *Illus.* 1931 (March, 1932).
- (24) Reimer, F. C. Value of Bordeaux mixture in blight control. *Trans. Oregon State Hort. Soc.* 17 (1925): 136-142. 1926.
- (25) ————— Pear blight control. Placer County (California) Fruit Growers' Convention 7th (1927): 76-81. 1928.
- (26) ————— Bordeaux spray for blight. *Oregon Agr. Exp. Sta. Bienn. Rept. Director 1928-1930*: 135-136. 1930.
- (27) Rosen, H. R. The relationship of different species of pomaceous hosts to the overwintering of the fire-blight germ. *Ark. Agr. Exp. Sta. 40th Ann. Rept. Bul.* 231: 68-70. 1928.
- (28) ————— The relationship of different species of pomaceous hosts to the overwintering of the fire-blight germ. *Ark. Agr. Exp. Sta. 41st Ann. Rept. Bul.* 246: 64-66. 1929.
- (29) ————— The life history of the fire-blight pathogen, *Bacillus amylovorus*, as related to the means of overwintering and dissemination. *Ark. Agr. Exp. Sta. Bul.* 244, 96 pp. *Illus.* 1929.
- (30) ————— Overwintering of the fire-blight pathogen, *Bacillus amylovorus*, within the beehive. *Science* 72: 301-302. 1930.
- (31) ————— Control of the blossom blight stage of fire blight. *Science* 76: 447-448. 1932.
- (32) —————, and Bleecker, W. L. Comparative serological and pathological investigations of the fire-blight organism and a pathogenic, fluorescent group of bacteria. *Jour. Agr. Res.* 46: 95-119. *Illus.* 1933.
- (33) —————, and Groves, A. B. Studies on fire blight: host range. *Jour. Agr. Res.* 37: 493-505. *Illus.* 1928.

- (34) Sackett, W. G. Some bacterial diseases of plants. Colorado Agr. Exp. Sta. Bul. 138, 23 pp. 1909.
- (35) Sherbakoff, C. D. The more important diseases of apples in Tennessee. Tenn. Agr. Exp. Sta. Bul. 145, 54 pp. Illus. 1932.
- (36) Stevens, F. L., Ruth, W. A., Peltier, G. L., and Malloch, J. R. Observations on pear blight in Illinois. Trans. Illinois Hort. Soc. 50 (1916): 216-227, 1917.
- (37) ————— and ————— Observations on pear blight in Illinois. Phytopathology 7: 75 (abstract). 1917.
- (38) Stewart, V. B. The fire blight disease of nursery stock. N. Y. (Cornell) Agr. Exp. Sta. Bul. 329: 316-371. Illus. 1913.
- (39) ————— and Leonard, M. D. Further studies in the role of insects in the dissemination of fire-blight bacteria. Phytopathology 6: 152-158. 1916.
- (40) Thomas, H. E. The longevity of *Bacillus amylovorus* (Burr.) Trev. in association with honey. Science 72: 634. 1930.
- (41) Tullis, E. C. Studies on the overwintering and modes of infection of the fire-blight organism. Michigan Agr. Exp. Sta. Tech. Bul. 97, 32 pp. Illus. 1929.
- (42) Waite, M. B. Results from recent investigations in pear blight. Proc. Amer. Assoc. Adv. Sci. 40 (1891): 315. 1892.
- (43) ————— The pear blight microbe. Trans. Peninsula Hort. Soc., 5th Ann. Session, pp. 32-34. 1892.
- (44) ————— The cause and prevention of pear blight. Yearbook U. S. Dept. Agr. 1895: 295-300. 1896.
- (45) ————— Pear blight and its treatment. Trans. N. Y. State Agr. Soc., Bur. Farmers' Institutes, Eastern and Western N. Y. Hort. Soc. 1897: 779-790. 1898.
- (46) ————— Fungus diseases of the apple and pear. Ann. Rep. Michigan State Hort. Soc. 27th (1897): 184-191. 1898.
- (47) ————— The relation of bees to the orchard. California Cultivator 18: 390-391. 1902.
- (48) ————— Pear blight work and its control in California. Fruit Growers Convention of California 31st. (1905): 137-155. 1906.
- (49) Waters, R. Fire blight—Incidence of the disease in New Zealand. New Zealand Jour. Agr. 24: 350-357. Illus. 1922.

- (50) Whetzel, H. H., and Stewart, V. B. Fire blight of pears, apples, quinces, etc. N. Y. (Cornell) Agr. Exp. Sta. Bul. 272: 32-51. Illus. 1909.
- (51) Woodcock, E. F., and Tullis, E. C. Extra-floral nectar glands of *Malus malus* and *Pyrus communis*. Michigan Acad. Sci., Arts, Letters 8: 239-243. Illus. 1927.

